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BENCH-SCALE TESTING OF BIODEGRADATION TECHNOLOGIES FOR PCBs IN NEW BEDFORD HARBOR (MA) SEDIMENTS

Radian Project No. 291-012-29-39

EBASCO Contract No. 4236-LAB-0062

FINAL REPORT

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June 2, 1989

ABSTRACT

This report presents the results of a bench-scale treatability study to test biodegradation as a potential treatment technology for remediation of PCB-contaminated sediments in New Bedford Harbor (Massachusetts). The study was conducted to support the evaluation of remedial technologies performed in the Feasibility Study for New Bedford Harbor. The study was designed to simulate conditions which may be expected in a full-scale biological treatment system to remediate New Bedford Harbor sediments.

The study consisted of two major phases, a biphenyl-degrading culture development step and a polychlorinated biphenyl (PCB) culture development phase. In both phases, the tests were conducted under aerobic conditions and with brackish media using shaker flasks. The standard procedure was to first acclimate and develop the culture using a fill and draw procedure, then confirm that successful development had occurred in a closed set of reactors. Microbial seed sources were obtained from New Bedford Harbor sediments and from PCB-contaminated sewage sludge. Sediments from two specific sources were used to test PCB degradation. One source contained relatively high concentrations of PCBs (>3,000 mg/l) and the second source contained lower concentrations of PCBs (<1,000 mg/l).

Biphenyl Culture Development

The biphenyl growth phase consisted of three substeps. In the first, a culture capable of biodegrading biphenyl was developed. Municipal sewage was used as the organic substrate, biphenyl was added, and the following six sources were used to seed the reactors: (1) New Bedford Harbor sediments with high levels of PCBs, (2) harbor sediments with low levels of PCBs, (3) harbor sediments from a combined sewer overflow (CSO) discharge point, (4) sediments from an intertidal zone contaminated with PCBs, (5) solids from a PCB aerobic digester (sewage sludge from Madison, Wisconsin), and (6) activated sludge mixed liquor.

After microbial growth in the acclimation reactors was confirmed by an increase in oxygen uptake rates and volatile solids, an initial set of confirmation reactors was initiated. The domestic sewage was eliminated and a small amount of seed from the first set of reactors was added. After an increase in turbidity was observed in these reactors (indication of growth), a second set of growth confirmation tests was conducted, using seed from the first set of confirmation reactors. Growth was observed in both the second and third sets of reactors, indicating that a biphenyl-degrading culture had been successfully developed.

PCB Culture Development

During the second phase of the study, a PCB-degrading culture was developed. The domestic sewage was replaced by the high and low PCB level harbor sediments (diluted to 1% solids) as the source of organic substrate. Seed from the biphenyl growth reactors as well as combined sewer overflow sediments, intertidal zone sediments, solids from a PCB aerobic digester, and activated sludge were added to the reactors. After 42 days of operation (3 retention times), a 10-day composite sample was collected and analyzed for PCB isomer groups (EPA Method 680). The results indicated approximately 15% overall reduction of PCBs in the high PCB-level sediment reactors and a 30% PCB reduction for the low PCB-level media. In tests with both sediments, the lesser chlorinated species (i.e., di- and trichlorobiphenyl) were degraded to a much greater extent than other isomer groups. PCB reduction was correlated to growth of biomass in only one set of reactors (the presumptive phase, high PCB-level sediment reactors).

A final set of reactors was operated to confirm that a PCB-degrading culture had been developed. The media again consisted of diluted harbor sediments (high and low level). After seeding (taken from the first set of PCB growth reactors) for the second time on the fifth day of operation, the reactor contents were sampled to establish the initial PCB concentration. The reactors were operated for 14 days without further addition of harbor sediments. At the end of two weeks, a final set of samples was collected. During the confirmation step, formaldehyde was added to two reactors to prevent biological growth. Any PCB reduction in these two reactors would be

an indication that volatilization or adsorption to the reactor walls had occurred. The test results were variable, however, as the formaldehyde apparently affected the amounts of PCBs which were released during the extraction phase of the analyses. In the high level reactors, overall PCB reduction (including all isomer groups) was similar to that observed in the control reactors, except that there was a preferential reduction in the concentration of "di" and"tri" isomer groups in the active reactors versus a uniform concentration reduction of all groups in the control reactors. In the low level sediment reactors, the removal efficiency was lower; dichlorobiphenyl was reduced 40-50%, but little or no removal of any other isomer group was noted.

Conclusions

The test results indicate that a microbial culture capable of degrading PCBs in a brackish water environment can be developed. However, the study results are not sufficient to prove the feasibility of biodegradation as a long-term remediation technology for PCB-contaminated New Bedford Harbor sediments. Only "di" and "tri" PCB isomer groups were degraded to a significant extent under conditions simulating those that would likely be utilized to treat large volumes of sediments. Dechlorination of PCB isomers which were not degraded in this study would probably enhance biodegradation rates. However, extensive research would be required to assess the potential for biologically mediated dechlorination as a pretreatment step to degradation of the PCBs.

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SUMMARY

Biphenyl Culture Development

1. The initial phase of the study indicated that a microbial culture capable of degrading biphenyl in a brackish environment was developed. This was shown by the growth of two generations of microbial communities in nutrient media with biphenyl as the only carbon source. The growth was indicated by an increase in turbidity and verified by microscopic examination.

PCB Culture - Presumptive Phase

1. Composite samples collected during this phase indicated that a reduction in PCB concentration was obtained in both the high and low PCB-level reactors.

2. Neglecting the results from reactor P1H, where only limited growth occurred, the overall PCB reduction ranged from 13-15% for the high level sediment reactors. For the low level sediments, overall PCB removal was 30%. There appeared to be a direct correlation between the amount of PCB reduction and the increase in total volatile solids level; this increase in microbial growth indicates that PCBs were being biologically degraded.

3. By isomer groups, the PCB reduction was greater for the lesser chlorinated species. For the high PCB-level sediment, dichlorobiphenyls were reduced 62-70%, trichlorobiphenyl 32-40%, and there was little or no removal of the higher chlorinated species. In tests with the low PCB-level sediments, some reduction in the "tetra" and "penta" levels was also noted, in addition to a slightly higher removal of the "di" and "tri"

isomer groups. The specific reduction by isomer groups for low level sediments was as follows: dichlorobiphenyl 79-82%, trichlorobiphenyl 48%, tetrachlorobiphenyl 14%, and pentachlorobiphenyl 6%.

PCB Culture - Confirmation Phase

1. During the confirmation phase, overall PCB reduction for the high PCB-level sediments ranged from 27-70%. The dichlorobiphenyl isomer group was reduced 83-100%, while the trichlorobiphenyls were reduced 64-87%. For the higher chlorinated groups, the reduction ranged from 0-7% in one reactor to 51-100% in another reactor. The reason for the wide range in percent removal of the higher chlorinated isomer groups is unknown.

2. In the low PCB-level sediment reactors, there was a reduction in dichlorobiphenyl of 39 to 50%, and little or no removal of any of the higher chlorinated groups. These reductions were substantially lower than that obtained for low level sediment during the presumptive phase.

3. The formaldehyde added to the control reactors to inhibit biological growth affected PCB analyses. In both control reactors, the initial PCB concentrations were approximately double that of the initial levels in the active reactors. The formaldehyde may have enhanced the release of bound PCBs during the extraction step of the analyses.

4. Without data from the control reactors, it is not possible to determine if any of the PCB removal observed was due to volatilization. Nevertheless, there was a nearly uniform reduction in concentration of each isomer group in the control reactors, whereas there was a preferential reduction of "di" and "tri" isomer group concentrations in the active reactors.

1.0 INTRODUCTION

1.1 Background Information

Extensive testing and study have shown the contamination of New Bedford Harbor (Massachusetts). The area north of the Hurricane Barrier (approximately 985 acres) is known to be underlain by sediments containing elevated levels of PCBs. PCB concentrations range from below the detection limit to over 30,000 ppm.

EBASCO Services Incorporated, Arlington, VA is under contract to the US EPA to perform a Remedial Investigation/Feasibility Study (RI/FS) on the New Bedford harbor site to assess the contamination and to evaluate alternatives for remedial action. The RI/FS Work Plan includes a detailed evaluation of detoxification/destruction technologies available to treat the contaminated sediments. A portion of this task is to conduct bench-scale treatability studies to determine if biodegradation would be potentially effective for use in decontaminating sediments. At the present time, the required PCB cleanup goals have not yet been determined. The bench tests described in this report were conducted to establish the feasibility of using aerobic biological treatment as a remedial technology for treatment of PCB-contaminated sediments in New Bedford Harbor.

1.2 Current Study

This report presents the results of a bench-scale program to develop a salt water microbial community capable of degrading PCBs. The project goal was to demonstrate this capability for treating PCBs sorbed to sediments from the Acushnet River Estuary at New Bedford, MA. The microbial community was developed from four different sources within the estuary as well as from sewage sludge containing PCBs. Instead of PCBs, biphenyl (the parent compound of PCB) was used as the carbon source for the initial culture development. A shift was then made to the actual PCB-contaminated sediments for the second phase of the study.

2.0 SCOPE OF WORK

The scope of work for this study was described in detail in earlier Radian documents prepared for this project. The revised Technical and Cost Proposal (July 23, 1987) and the Experimental Project Plan (October 5, 1987) are presented as Appendix A and B, respectively. Modifications that were made to the original scope are discussed in this section.

2.1 Selection Techniques

The selection technique was designed to develop a complete microbial community adapted to degrading PCBs in the presence of other substrates and microorganisms. This is in contrast to simply isolating a microbial population capable of degrading PCBs, the process more often used in developing an enriched culture. The open culture procedure was chosen for three main reasons. First and most importantly, it would be prohibitively expensive to prevent the introduction of alien microorganisms in a full-scale treatment unit. Thus, it is important that the microbial population be able to compete with other microorganisms. Second, since there are numerous substrates which can serve as energy sources in the bay sediments, it is important that the population selected be able to degrade PCBs in the presence of other substrates. Third, it may be important to have alternative energy sources available, particularly those which are analogs to PCBs and would require a similar enzyme system for degradation. Alternative energy sources would be important if microorganisms cometabolize PCBs (i.e., use other substrates as the energy source), as has been suggested in the literature.

The culture was developed using six sources of microorganisms capable of degrading PCBs. Multiple seed sources were used to enhance the probability of introducing organisms that had naturally developed the ability to degrade PCBs in an environment similar to New Bedford Harbor.

The following four sources of microbes were obtained from the estuary within the New Bedford Harbor area.

1. Heavily contaminated sediments from the area adjacent to the Aerovox facility;

2. Moderately contaminated sediments from an area approximately 600 feet north of Aerovox;

3. Sediments collected adjacent to a CSO (combined sewer overflow) outfall near Coffin Street; and

4. Sediments from an intertidal zone approximately 500 feet north of Aerovox.

It was anticipated that the slightly contaminated sediments would contain the most desirable microbial population. These microbes should have adjusted over time to the presence of low level PCBs without being shocked by high PCB concentrations and without competition from diverse populations. Sediments from a high PCB concentration area were used as another source of microbes adjusted to PCBs. Both the high and low concentration PCB seed samples were collected from the top several inches of sediment to obtain an active aerobic culture.

A culture from near a combined sewer overflow outfall in the estuary was used as a source of seed that had been exposed to numerous organic compounds and frequent introduction of microbes. The purpose of using this seed material was to introduce a diverse microbial population which could potentially contribute microorganisms capable of degrading PCBs.

Microorganisms from a nearby intertidal zone were also added to the culture to diversify the population. These organisms were acclimated to a relatively stressful environment. For example, the intertidal zone is exposed to cyclical wet and dry periods.

In addition to these four saltwater sediment sources, two additional seed samples were used. These two were from a freshwater environment; however, many of the organisms within the population can withstand varying degrees of salinity. The two sources were:

1. Solids from a functioning PCB aerobic sludge digester containing PCB-contaminated sewage sludge from Madison, Wisconsin; and

2. Samples of activated sludge microorganisms (mixed liquor from the South Shore Treatment Plant, Milwaukee, Wisconsin).

It was initially intended to use a microbial culture from a commercial source or research laboratory as a seventh seed source. Attempts to locate a culture capable of competing effectively in a saline environment were unsuccessful during the time frame between project approval and startup.

For the initial cultures, biphenyl was used as the carbon source instead of PCBs, primarily because PCB-degrading enzyme systems that accomplish ring fission can generally also degrade biphenyl. However, the reverse is not always true, all enzyme systems that degrade biphenyl may not be capable of degrading all PCBs. Therefore, after initial tests using biphenyl, PCB-contaminated sediments from New Bedford Harbor were used to verify the suitability of the culture for PCB degradation. In adding harbor sediments as a source of PCBs, additional seeding of microbes with potential PCB degradation capabilities was also accomplished.

2.2 Culture Development

The development of the enriched culture was performed in two steps. In the first step, a culture was established which was able to degrade biphenyl in the presence of other substrates. Using the first culture as seed, a second step was undertaken to develop a PCB-degrading culture using actual contaminated sediments as the carbon source. Further detail on development of the biphenyl and PCB cultures is provided in the following sections.

2.2.1 Biphenyl Culture Development

The biphenyl cultures were developed by adding biphenyl to a mixture of salt water and raw sewage. The media was initially seeded with each of the six microbial seed sources. A wasting schedule was established to maintain a

hydraulic retention time of five (5) days. General growth in the reactors was monitored by determining the total solids, total volatile solids, and oxygen uptake rates.

Biological biphenyl degradation was demonstrated by adding seed from the sewage/biphenyl cultures to a set of reactors containing nutrient enriched salt water and biphenyl. Biphenyl degradation and corresponding biomass growth was monitored by measuring the increase in the turbidity of the nutrient media of these verification cultures. To ensure that the turbidity was due to growth from biphenyl degradation and not from use of other substrates introduced with the seed, a second set of growth confirmation cultures (with biphenyl as only carbon source) was developed using the first set as the seed source.

2.2.2 PCB Culture Development

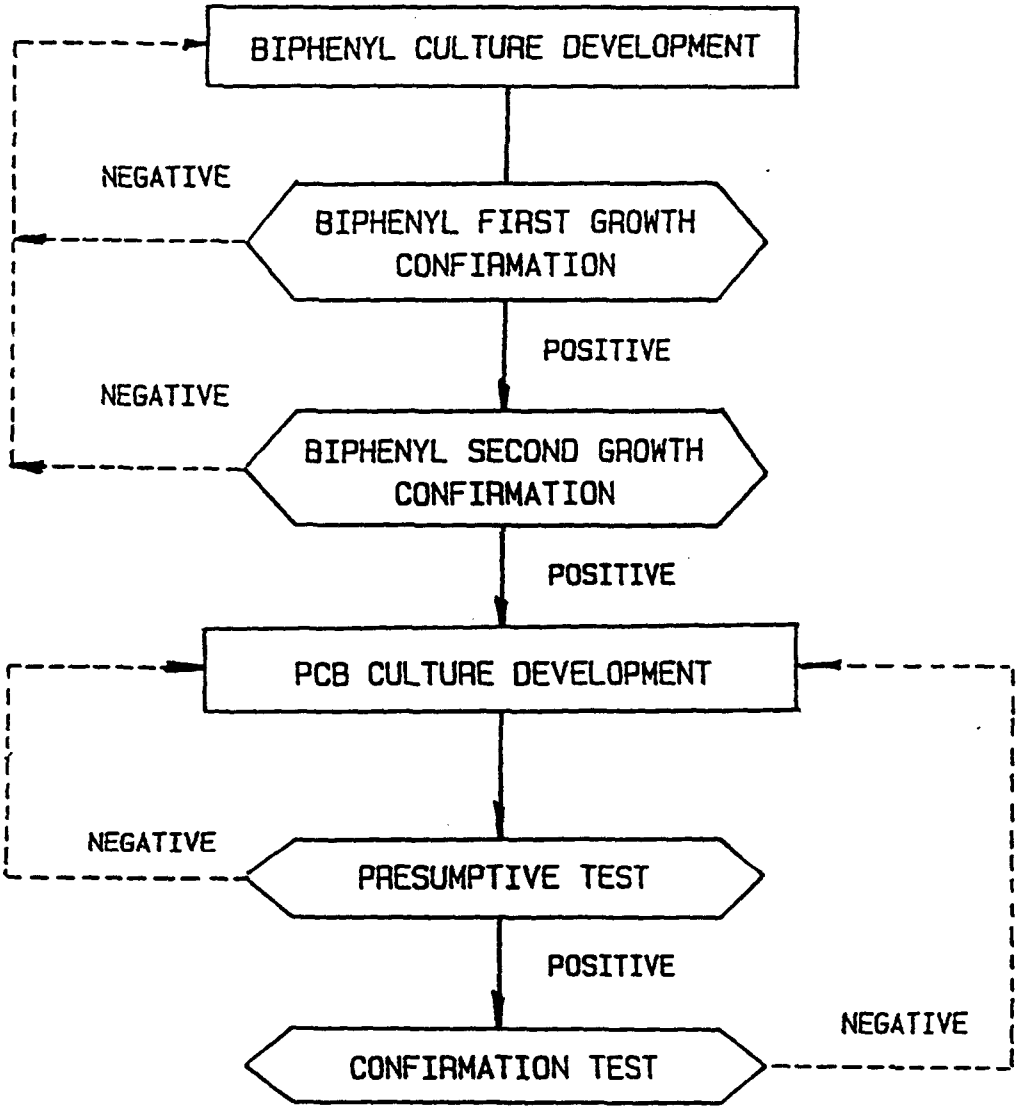
The sediment PCB cultures were developed following a similar process as the biphenyl cultures; however, sediment from the New Bedford Harbor was used instead of the raw sewage. Biphenyl was again added to each reactor as a potential cometabolite and a protected substrate source to boost the populations previously enriched. Seed was taken from the biphenyl/sewage cultures and from the other six microbial sources previously discussed. General growth in the reactors was monitored based on solids production and oxygen uptake rates.

After a period of time sufficient to reach steady state conditions, the ability of the culture to degrade PCBs was determined in a two-step process consisting of presumptive tests and confirmation tests. In the presumptive test, a ten-day composite sample of the feed and effluent from each reactor was collected and analyzed for PCB content. Differences in feed and effluent PCB concentration were used to compute PCB loss. The overall PCB removal could not, however, be attributed entirely to biodegradation. Mechanisms such as volatilization or sorption to the reactor walls could also have occurred.

In the confirmation tests, PCB biodegradation was confirmed by adding seed from the sediment cultures to reactors containing a fresh mixture of preaerated contaminated sediment, biphenyl, and nutrient enriched salt water. Preaerated sediment was used to prevent death of the culture due to oxygen depletion from the initial oxygen demand of anaerobic sediments. In addition, control flasks were set up containing the contaminated sediments and seawater, but also with sufficient biocide to inhibit any biological growth. Differences between PCB removal observed in the confirmation test reactors and the control reactors were used to assess the degree of PCB biodegradation in the confirmation reactors.

The overall project sequence and decision points are illustrated in Figure 1. At a number of milestones, decisions were made about whether to advance to the subsequent step or repeat a previous culture development step. This approach was designed to maximize the potential of successfully developing a PCB-degrading culture in an environment similar to that in New Bedford Harbor.

FIGURE 1
MAIN PROJECT FLOW DIAGRAM



3.0 TEST PROCEDURE

A detailed discussion of the procedures and equipment used during this project is presented in this section. The project involved six sets of reactor tests; this included three sets of biphenyl cultures and three sets of PCB cultures. In this section, the following topics are described for each set of reactor testing:

- The media used for culture development;
- Initial reactor conditions;
- Reactor operation;
- Sampling and analysis; and
- Milestones and decision points.

The study also included operation of a set of control reactors in which biological activity was inhibited by formaldehyde addition. A description of the sterilization procedure used in conjunction with the control reactors is also provided in this section.

Quality assurance standards were maintained throughout the course of the study. The standards were established in the Quality Assurance Project Plan (QAPP) initially prepared by Radian on January 20, 1988, and revised and issued in final form on April 22, 1988. Quality assurance procedures are not addressed in this section; however, the project QAPP appears in Appendix C.

3.1 Biphenyl Cultures

The bench-scale development of biphenyl cultures involved acclimation of the microbial culture, and confirmation that the developed culture was using biphenyl as a carbon source.

3.1.1 Acclimation of the Microbial Culture

The goal of the acclimation phase was to allow development of a culture capable of degrading biphenyl in the presence of alternative substrates. This acclimation period allowed for shifts in the microbial

population species and optimization of microbial metabolisms. For example, this period allowed microorganisms not previously exposed to biphenyl to adjust to its presence. In addition, those microbes already capable of using biphenyl were allowed to adjust to the quantity of biphenyl-degrading enzymes produced to correspond to the availability of biphenyl. This allowed optimization of biphenyl degradation and resulted in the biphenyl-degrading species being able to compete successfully with other microorganisms in the culture. The conditions maintained in the biphenyl acclimation cultures are discussed below.

3.1.1.1 Media

The media used in this culture development was raw sewage (obtained from the Milwaukee Metropolitan Sewerage Commission, South Shore Plant) with the addition of sea salts (trade name "Instant Ocean" manufactured by Aquarium Systems) to increase the salinity of the solution. In addition, the following micronutrients were added as supplements:

$MgSO_4.7H_2O$ (0.05 g/L)
$FeSO_4.7H_2O$ (0.01 g/L)
$CuSO_4.5H_2O$ (0.01 g/L)
$CoCl_2.6H_2O$ (0.01 g/L)

The micronutrients were added to each batch (typically 3,000 mls) of raw sewage. This stock solution was used to initially fill the reactors, and for the daily additions. Although this solution was refrigerated between use, that portion which was to be added to the reactors was allowed to warm to room temperature to prevent shocking the reactors.

A sample of seawater was collected from the estuary approximately 300 feet north of Aerovox, and found to have a specific conductivity of 4,600 umohs. Sea salt was added to the raw sewage stock solution to match this conductivity. Analysis of the reactor contents indicated this conductivity corresponded to a salinity of approximately 3 g/kg.

Biphenyl was added to the reactors on a daily basis. It was not added as a constituent of the media because of the potential for volatilization. Biphenyl has a soluble half life in quiescent water in the range of two to six hours. The biphenyl was ground into fine particles prior to addition.

3.1.1.2 Initial Conditions

For the initial series of tests, six reactors were used. The initial reactors, numbered 1 through 6, were placed into operation on February 22, 1988. Each reactor consisted of a 500-ml Erlenmeyer flask, with a rubber stopper wrapped with aluminum foil to prevent sorption. Air was circulated in the head space of the reactor via two glass tubes extending through the stoppers. The tubes were placed well above the liquid surface to prevent stripping of any compounds from the liquid phase. The air flow rate was set at approximately 100 ml/min. The air flowed from one reactor to the next through flexible tubing. The reactors were shaken at a rate sufficient to ensure aerobic conditions in the media.

The initial contents of each reactor were 150 mls of media, 0.1 grams biphenyl, and seeds from each of the six microbial sources identified and discussed in Section 2. To seed each reactor, 0.5 grams of sediment (wet weight) from each of the four harbor sources and 1 ml of seed from the other two sources were added. The harbor seed sediments were slurried to make it easier to add a uniform amount to each reactor.

3.1.1.3 Reactor Operation

After the initial setup, the reactors were placed on a Burrell wrist arm shaker which was operated continuously. The flasks were fed on a daily basis, 6 days a week, for the duration of the acclimation phase. The reactors were operated in a batch fill and draw mode. Each day the flasks were weighed and any evaporative losses were replaced with tap water. Tap water was used since dissolved solids buildup was not considered to be a problem during

this phase of the study. To feed the reactors, the media was allowed to settle for approximately 15-30 minutes. A 30 ml volume of supernatant was then removed and replaced with 30 ml fresh media and 0.1 grams of biphenyl. On every sixth day of operation, twice the volume (60 ml) of media and biphenyl were exhanged to provide a two-day food supply since the reactors were not fed on Sundays.

The removal and addition of 30 ml on a daily basis resulted in a five-day hydraulic retention time. This retention time was selected to provide for a sufficient quantity of readily available organic food source in the sewage for the microorganisms and removal of metabolic byproducts which might inhibit continuous growth. Another reason for selecting a five-day retention time was to ensure that the food supply would be sufficient to allow for survival of the microbes until they could shift to biphenyl as a food source, but low enough to prevent continuous growth on the organics in the sewage.

As previously mentioned, the salinity of the seawater from the area where the sediment samples were collected was found to be approximately 3 g/kg. This salinity was much lower than would be expected for water in the southern portion of the estuary. The low salinity was probably the result of a combination of factors including the location of the collection point relative to the Acushnet River discharge, the ice and snow melt that was occurring at the time of collection, and the discharge of cooling water from a nearby manufacturing plant. Published values of salinity for water samples taken at Wood Street Bridge and Coffin Cove indicate that salinites may range from <1 g/kg at the Wood Street Bridge area to approximately 30 g/kg in the southern part of the estuary near Coggeshall Street Bridge. To more closely simulate the conditions which would be expected in most of the estuary and harbor, the decision was made to increase the salinity by 10 g/kg. This was done by adding sea salt in increments of 2 g/kg beginning on March 1 (Day 9) and ending on March 5 (Day 13).

On Day 5 (February 26), all six acclimation reactors were reseeded using the procedure originally used in setting up the units. The purpose of reseeding the reactors was to expose newly introduced microorganisms to a

substrate matrix depleted in easily biodegradable organics and enriched in biphenyl. In the initial seeding, the raw sewage substrate provided a high concentration of readily degradable organics. After five days of operation, the concentrations of these species should have been much lower. By reseeding at this point, the biphenyl degrading species were given a competitive advantage over those species that primarily use more easily degraded organics as a carbon source.

At the completion of the biphenyl acclimation phase, the contents of reactors 2 and 3 and reactors 4 and 6 were combined and placed in 1,000 ml flasks (designated as 7A and 8A, respectively). These flasks were maintained and monitored throughout the remainder of the program. The same fill and draw sequence was followed with the exception that twice as much media (60 ml) was transferred daily to maintain the 5-day hydraulic retention time.

3.1.1.4 Sampling and Analysis

Solids analyses and oxygen uptake rates were conducted twice a week to monitor microbial growth in the biphenyl/sewage mixture. When total and total volatile solids samples were collected, the reactor contents were not allowed to settle. While being agitated, 30 ml were removed and replaced with 30 ml of media. Oxygen uptakes were also performed twice a week. Because 300 ml of media is required to perform an oxygen uptake test, the contents of two reactors were combined for this analysis. After the test, the sample was redivided into 150 ml fractions and returned to the culture reactors. Mixing pairs of flasks also allowed cross seeding of cultures, increasing the opportunity for successful acclimation. Reactor pairs to be mixed were selected randomly for each sampling period. The oxygen uptake analysis was made approximately 3 hours after the biphenyl and media were added.

3.1.1.5 Milestones and Decision Points

The key decision point in the biphenyl acclimation phase was indication of microbial growth. For example, increase in the percent volatile solids or in the oxygen uptake rate are growth indications. At five-day increments, the data were reviewed and the decision was made to either reseed

the reactors or to continue. A major decision point was scheduled at approximately Day 15, when three hydraulic retention times had been completed. Statistically, 90% of steady state physical conditions should have been achieved after three retention times. As will be discussed in Section 4, substantial growth was observed at this point and the next phase of the program was initiated on March 9 (Day 17).

3.1.2 Biphenyl Culture Confirmation

To determine if biphenyl degrading microorganisms were present and competing successfully with other microorganisms in the acclimated phase, it was necessary to test the culture's ability to grow in a medium with biphenyl as the sole carbon source. This was done in the biphenyl culture confirmation phase. The approach used was to add 1 ml of seed from the sewage/biphenyl acclimation culture to media composed of seawater, nutrients, and biphenyl, and to measure turbidity over a five-day period. Increases in turbidity were interpreted as either microbial growth or microbial degradation of biphenyl. It has also been found that accumulation of one of the degradation products of biphenyl induces a yellow color to the media; therefore, the appearance of yellow color was also documented as a potential growth indication. Seeding and growth was then repeated in a second set of growth confirmation reactors using the existing cultures as the new seed. This was done to ensure that biphenyl was being used as the primary carbon source for biological growth.

3.1.2.1 Media

The media used in the biphenyl confirmation tests was made from organic carbon-free water (deionized), sea salts, the micronutrients previously listed, and the following nitrogen and phosphorus sources:

K_2HPO_4 (1.3 g/L)
KH_2PO_4 (0.82 g/L)
$(NH_4)_2SO_4$ (1.0 g/L)

Phosphorus and nitrogen were added to ensure that biological growth was not limited by noncarbon sources. Also, these forms of the nutrients provided a pH buffer in the range optimal for most microbial growth. The prepared media was filtered through Whatman #1 filters to remove any gross undissolved material before use in the reactors.

3.1.2.2 Initial Conditions

Six reactors were prepared (500-ml Erlenmeyer flasks) and filled with 150 ml of media and 0.1 grams of biphenyl. The flasks were covered with aluminum foil to limit the light available to the culture. Seeding of the first set of reactors (first growth) was performed by transferring 1 ml of seed from the sewage/biphenyl acclimation reactors to the growth confirmation reactors (combination of acclimation reactors 2 and 3 to seed growth reactors 1A and 2A, 7 and 5 to seed 3A and 4A, and 4 and 6 to seed 5A and 6A). After seeding, the flasks were stoppered, the air flow tubing attached, and the flasks placed on the Burrell shaker. Operation of this phase of the program began on March 9 (Day 17).

The second set of growth confirmation reactors were set up similar to the first with the exception of the seed. The seed in this case was taken from the first growth reactors. The second growth reactors were started on March 15 (Day 23). For each set of reactors, a blank was also prepared. This reactor contained the media and the biphenyl, but no seed was added.

3.1.2.3 Reactor Operation

The reactors were operated in a batch mode (i.e., media was not added or removed during the test). Biphenyl (0.05 grams) and deionized water were added each day to replace evaporative losses. Prior to the addition of the biphenyl, the reactor contents were checked. If any undissolved biphenyl remained, the amount added was reduced. On some occasions, biphenyl addition was not required because of high residual concentrations in the reactors.

3.1.2.4 Sampling and Analysis

The turbidity of each of the reactors was measured six times a week. The sample used to determine turbidity was returned unchanged to the reactor from which it was collected. Turbidity measurements were made before the addition of biphenyl to eliminate undissolved biphenyl as an interference. The reactors were also checked each day for the onset of foaming or changes in the color. Both of these conditions were checked as possible indications of biphenyl degradation.

3.1.2.5 Milestones and Decision Points

The critical times in biphenyl confirmation testing were on Day 5 and 10 of each test set. On the fifth day (Set No. 1), the turbidity data were reviewed. At this point, all reactors had shown an increase in turbidity as well as a change in color (clear to yellow). The decision was then made to proceed to the second growth confirmation phase. There was some concern that the transfer of seed to the second set of reactors may have occurred slightly past the optimum time (Day 6 versus Day 5). As a means of confirming the results, two additional reactors were started on March 15 (7R and 8R).

For the second set of growth confirmation reactors, the decision points observed were similar to those noted during operation of the first set of reactors. After approximately five days of operation, the turbidity values were evaluated. A significant increase was observed; therefore, the next phase of the program (PCB Culture Development) was initiated.

3.2 PCB Culture

The goal of this task was to acclimate the developed culture and demonstrate the ability of this culture to degrade PCBs. The development and testing of PCB-degrading cultures proceeded in the following stages:

A. Culture Acclimation
B. Presumptive Testing
C. Confirmation Testing

After an acclimated culture was produced, the development of a PCB-degrading culture was initiated. The sewage/biphenyl acclimation culture was used as the seed for this step. The culture was developed using actual sediments from New Bedford Harbor supplemented with micronutrients, nitrogen, phosphorus, and biphenyl.

3.2.1 Culture Acclimation

Because of the change in carbon sources and the change in reactor operating parameters, it was necessary to reacclimate the sewage/biphenyl acclimation culture to the sediment/biphenyl mix.

The change in carbon sources was a result of shifting from a sewage to a sediment substrate. In sewage, most of the carbon sources are soluble organics and are readily degradable. In sediments, most of the carbon sources are relatively insoluble and are sorbed to solids. The microbial community capable of degrading sewage substrates may not contain the same population levels or distribution of microbes as would be found in the sediment substrate.

Operating conditions were changed to a 14-day solids and hydraulic retention time; this may also have affected the microbial population. This change resulted in a different food/microorganism ratio as well as providing a longer time period to degrade the substrates. The increase in retention time was made because prior studies had shown that microbes degrade PCBs much slower than biphenyl. Fourteen days was selected as a retention time that should allow significant PCB degradation.

3.2.1.1 Media

The sediments in the media used in this phase of the project were taken from two sources within the estuary; these were the PCB hot spot adjacent to the Aerovax facility (>3,000 mg/l PCB) and a less contaminated area approximately 600 feet north of Aerovox (<1,000 mg/l PCB). Sediments with two quite different PCB levels were selected to provide more information on the limits of the biodegradation process. Radian was supplied with two

separate samples of each sediment source. One sample was used as media and the second as seed material (Section 2.1). Both sediments were diluted to 1% solids (dry weight basis) using actual seawater collected from the harbor. Micronutrients (see Section 3.1.1.1), nitrogen, phosphorus (see Section 3.1.2.1) and sea salts (10 gr/kg) were also added to the media. The media solutions were prepared in 2- to 3-liter batches and refrigerated between use.

3.2.1.2 Initial Conditions

Seven reactors (1-liter Erlenmeyer flasks) were prepared for the PCB culture acclimation tests. Each reactor was filled with 300 ml of media and 0.2 grams biphenyl. The increase in reactor size and media volume made it possible to collect larger samples for analysis. Four of the reactors were filled and fed media prepared with the high concentration sediments, and three were prepared with sediments from the less contaminated area.

After filling, the reactors were placed on the Burrell shaker and agitated continuously. For this phase, the flasks were stoppered and vented, but air was not forced through the head space. During the biphenyl culture development phase, problems were encountered with condensation in the interconnecting air lines. By removing the airlines during this phase, these problems were eliminated. Several checks during the first week of operation indicated that sufficient dissolved oxygen was being maintained without the forced air circulation.

The reactors were seeded with 10 ml of culture taken from the sewage/biphenyl acclimation reactors (5 ml from 7A and 5 ml from 8A), and 1 ml of seed from each of the following sources (described in Section 2.1):

1. CSO sediment;
2. Intertidal zone sediment;
3. Municipal activated sludge; and
4. Sludge from a functioning PCB aerobic digester.

The seven reactors were started on March 22, 1988. Reactors 1H through 4H contained the high level PCB sediment and 5L through 7L the low level sediment. Seed was added on March 23, 25, 28, and 30, and April 1. On May 3, it was determined that the low level media had been prepared incorrectly (it was actually prepared using high level sediment). Two additional reactors were then started with low level sediments (5L' and 6L'). Seeding occurred on May 4, 6, 9, 11, and 13. In seeding the second set, 1 ml of seed material was taken each time from the original low level reactors in addition to the other four seed sources.

3.2.1.3 Reactor Operation

The reactors were operated in a fill and draw mode similar to that used for biphenyl culture acclimation. The one exception was that the reactor solids were not allowed to settle prior to withdrawal. Each day the reactors were weighed and evaporative losses were made up by adding distilled water. Twenty-two milliliters were withdrawn daily, resulting in a hydraulic retention time of 14 days. The volume withdrawn was replaced with the 1% solids media and biphenyl. The reactors were operated for 41 days before proceeding to the next phase.

Although the objective of this phase of the study was to develop a PCB-degrading culture, small amounts of biphenyl continued to be added. The biphenyl was added as a stimulant and to aid in maintaining the culture. Ideally, the microbes would quickly degrade the biphenyl, then be sustained for the remainder of the day by degrading PCB material. Initially (first 5 days) 0.05 grams of biphenyl was added daily. This dose was then cut back to 0.01 grams in an attempt to ensure that the concentration of biphenyl was insufficient to completely sustain the microbes. After three weeks of operation with relatively slow growth in the reactors, the amount of biphenyl added was increased to 0.02 grams daily for the remainder of the program.

3.2.1.4 Sampling and Analysis

General operating variables were monitored, including solid levels, oxygen uptakes, and dissolved oxygen levels. Both total and total volatile

solids were analyzed twice a week using the effluent from the fill and draw operation. Dissolved oxygen levels were also measured twice a week and oxygen uptake rates were performed once a week.

3.2.1.5 Milestones and Decision Points

Decision points for the PCB acclimation phase were scheduled every 14 days. Since some growth was observed, the operation of the reactors was continued. After 41 days of operation, the next phase (presumptive testing) of the program was initiated.

3.2.2 Presumptive Testing

Presumptive testing involved measuring the change in the mass of PCBs in the sediment reactors over a 10-day period. The test was considered presumptive in that it measured only the change in PCB mass, and did not demonstrate that the disappearance resulted from biodegradation. The presumptive tests involved simply continuing the operation of the PCB acclimation reactors for a 10-day period beyond the initial 41 days. The presumptive testing began on May 2 for the high PCB-level sediment reactors (1H through 4H) and on June 14 for the low PCB-level sediment reactors (5L' and 6L').

3.2.2.1 Sampling and Analysis

During presumptive testing, samples were collected and analyzed for total solids and total volatile solids; samples were also collected for analysis of total PCB levels and distribution of PCB congeners in both the reactor feed and effluents. A 10-day composite of reactor feed was prepared. Twenty-two ml of media were composited each day in an amber bottle and refrigerated. To determine the corresponding information in the reactor effluents, a set of effluent composite samples was also collected for analysis. Again, 22 ml of media were collected from each reactor, placed in separate composite sample bottles, and refrigerated. Total solids and total volatile solids analyses were performed on samples from high PCB-level sediment reactors only.

PCB analyses were performed by Gulf South Research Institute (GSRI). The samples were analyzed for pesticides/PCBs by gas chromatography in accordance with the protocols specified by USEPA Method 608. Samples were also analyzed by gas chromatography/mass spectrometry in accordance with USEPA Method 680 to determine the PCB isomer groups. PCB analyses were not included in the Quality Assurance Project Plan; as a result, QA/QC data for analyses conducted by GSRI were not provided to Radian during the study.

From each composite sample, four individual 25-ml samples were collected and placed in amber I-Chem Series 300 bottles. This provided an individual sample for each method of analysis, and a sample for duplicate analysis, if necessary. These samples were packaged and sent from Radian to E. C. Jordan (Portland, ME); E. C. Jordan then submitted samples to GSRI. In addition, select samples were sent to Radian Corporation laboratories (Austin, TX) for PCB analysis by Method 680.

3.2.2.2 Milestones and Decision Points

The initial intent was to decide on a course of action based on PCB concentration analyses. If a reduction in PCB concentration was found, and the reduction was greater in the lower chlorinated biphenyls than overall, the confirmation phase would be initiated. Alternatively, if no significant change was observed, a second set of composite samples would be taken. Due to delays involved in getting the samples analyzed, this review process was not possible. At the end of the 10-day presumptive test, the confirmation phase was initiated.

3.2.3 Confirmation of PCB Biodegradation

A set of confirmation reactors was operated to determine if PCBs were actually biodegraded and not sorbed to the reactor walls or volatilized; these batch reactors were seeded from the presumptive phase cultures. Two control reactors in which biological growth was eliminated were also operated in this phase. Evidence for biological decomposition was sought by determining if reduction in PCB levels was greater in the biologically active reactors than

in the control reactors. In addition, differences in the distribution of PCB isomers in effluents from the biological reactors and the control reactors were measured and used as another indication of biological degradation.

3.2.3.1 Media

The media used in the confirmation reactor tests were the same as that used in the acclimation and presumptive phase reactors. It consisted of the harbor sediments diluted to 1% solids, micronutrients, sea salt, and nitrogen and phosphorus sources.

3.2.3.2 Initial Conditions

Eight 1-liter flasks were used as confirmation reactors; six were used for biological degradation and two as controls. Of the eight, five contained high PCB concentration (i.e., >3,000 mg/l) sediment media (C1H, C2H, C3H, C4H, C7H) and three contained the lower PCB-level (i.e., <1,000 mg/l) material (C5L, C6L, C8L). A biological toxin, paraformaldehyde, was added to the control reactors (C7H and C8L) to prevent biological growth (see Section 3.3).

The reactors were initially seeded after 24 hours of aeration. Preaeration was conducted to ensure that the reactor media was aerobic before introduction of the seed. Ten milliliters of seed culture were added to each of the biological reactors. The seed was taken from the presumptive test reactors (1H to C1H, 2H to C2H, 3H to C3H and C7H, 4H to C4H, 5L to C5L, 6L to C6L and C8L). Seeding was repeated at the beginning of Day 5 to enhance the probability of successful PCB degradation.

Key milestones in the PCB degradation confirmation phase are listed below:

	High Level Media		Low Level Media	
	Date	Day	Date	Day
Setup	May 12	81	June 23	123
Initial Seeding	May 13	82	June 24	124
Second Seeding	May 17	86	June 28	128
Completion	May 31	100	July 12	142

3.2.3.3 Reactor Operation

The confirmation reactors were agitated for a period of 14 days after the second seeding. No drawing or filling took place during this period. Biphenyl was added to the reactors each day (0.02 grams); when evaporative losses totaled 10 grams, distilled water was added to make up for the loss.

3.2.3.4 Sampling and Analysis

The confirmation reactors were sampled for PCBs and solids at the beginning and end of the confirmation period. Initial samples were collected after the second seeding. The seed was added and the reactor contents mixed for 30 minutes before sample collection. As during the presumptive phase, four individual samples were collected from each reactor and placed in I-Chem Series 300 bottles. PCB analysis by both methods (608 and 680) was again performed by GSRI. Selected samples were also sent to Radian Corporation for PCB analyses by Method 680.

The initial volume in each reactor including seed was 420 ml. After removing 120 ml for the initial sampling, the reactor volume was maintained at 300 ml for the remainder of the confirmation period.

After 14 days of operation, the final set of samples was taken from each reactor and sent to GSRI and Radian for PCB analyses.

3.3 Sterilization Procedure

To determine the mechanism of PCB removal in the confirmation reactors, it was necessary to operate a control reactor in which no biological activity would occur. In this study, a set of control reactors was sterilized by adding formeldehyde.

During the culture acclimation substep (Section 3.2.1), an extra reactor (7L) containing low PCB-level sediment was prepared and operated similarly to the other six. After approximately three weeks of operation, a sample was taken from this reactor and formaldehyde sterilization tests were performed to determine the optimum concentration necessary to achieve culture sterilization. In this procedure, low PCB-level media were dispensed in 50-ml aliquots into 500-ml Erlenmeyer flasks; the media was then treated with a 37% formaldehyde solution to obtain desired final formaldehyde concentrations in the 0-10% range.

The sterilization test flasks were placed on an orbital shaker at approximately 100 rpm and were maintained at 20°C for a period of 14 days. At the end of the 14-day incubation period, total plate counts for aerobic and facultative anaerobic bacteria were conducted on the contents of each flask. The plate count method used was similar to the method described in "Standard Methods for the Examination of Water and Wastewater," AWWA, APHA, WPFC, 16th Edition, 1985. The culture medium used in the plate count was Marine Agar 2216 prepared from reagent chemicals and nutrient agar. Each flask was plated using four dilutions in triplicate in the range from 10^{0} to 10^{-5}. The results of the plate counts are presented in Table 1.

TABLE 1. TOTAL PLATE COUNTS RESULTS FROM FORMALDEHYDE STERILIZATION TESTS

Test Flask	Formaldehyde Concentration	Total Plate Count CFU/mL[a]
1	0%	2.2 E+05
2	1	<1
3	3	<1
4	5	<1
5	5	<1
6	10	<1

[a]CFU/mL - Colony forming units per mL.

Based on the test results, a formaldehyde concentration of 2% by weight was selected for confirmation test control reactors to ensure a sterile environment.

When the confirmation phase control reactors were set up, paraformaldehyde was added to achieve a dosage of 2% by weight. The formaldehyde concentration was monitored in the high PCB-level control reactor on a daily basis using the Hantzsch Reaction Method given in Formaldehyde, Walker, 3rd Edition, p. 472. This is a spectrophotometric method based on the reaction of formaldehyde with acetylacetone and an ammonium salt to give diacetyldihydrolutidine. Reactor formaldehyde concentration and corresponding QC results are presented in Table 2.

A blank, standard, spike, and a duplicate of the control reactor contents were analyzed daily. The blank consisted of deionized water to which the reagents had been added, the standard was a prepared solution which contained 5.41 mg/l of formaldehyde, and the spike sample consisted of the reactor contents to which a known amount of standard solution was added. For each set of analyses, the recovery was calculated based on the following equation:

TABLE 2. CONCENTRATION OF FORMALDEHYDE IN PCB CONFIRMATION PHASE HIGH PCB-LEVEL CONTROL REACTOR

Date	Day	Blank mg/l	Std. 5.41 mg/l	Reactor Contents %		Recovery[a] %
5-13	82	0.11	5.35	1.14	1.24	93.6
5-14	83	0.10	4.84	1.30	1.44	98.8
5-16	85	0.18	4.82	2.77	2.47	87
5-17	86	0.12	5.29	OR[b]	OR	ND[c]
5-18	87	0.10	4.86	3.92	3.44	106
5-19	88	0.20	5.30	1.52	1.50	100
5-19	88	0.06	5.24	OR	2.44	100
5-20	89	0.06	5.26	2.08	2.05	96.5
5-23	92	ND	ND	2.38	2.38	102
5-24	93	0.06	5.36	2.12	2.19	103
5-25	94	0.06	5.20	2.20	2.22	99.6
5-26	95	0.03	5.30	2.17	2.17	102
5-27	96	-0.06	5.24	2.37	2.40	100.8

[a]Recovery equals 100 (M-C)/A where: M = concentration in spiked sample, C = concentration in original sample, A = theoretical increase in concentration due to spike.

[b]OR - Outside of the range of the calibration curve that was prepared.

[c]ND - Not determined.

$$\% \text{ Recovery} = 100 \left(\frac{M-C}{A}\right)$$

Where M = Concentration of formaldehyde in the the spiked sample (corrected for dilution).

C = Concentration of formaldehyde in the sample.

A = Known concentration of formaldehyde added by spiking (corrected for dilution).

No formaldehyde analyses were performed for the low PCB-level control reactor.

Initially, some variability in the results was obtained because of improper sampling technique. The formaldehyde appeared to react with, or adsorb to, the sediments. For instance, when the supernatant was sampled (May 12 and 13), a low formaldehyde concentration was obtained. After this trend was noted and the sampling technique modified to obtain a more representative sample, reproducible results were obtained. On May 19, a uniform sample was apparently not obtained, and a low formaldehyde concentration was measured. Another sample was taken, and as shown in Table 2, more representative results were obtained. The results in Table 2 show that the formaldehyde concentration remained approximately the same throughout the 14-day period. As a result, it was not necessary to add formaldehyde after the initial dose.

On May 26 (14 days after startup), a sample was taken from the high PCB-level control reactor for a total plate count determination. The results indicated there was less than 1 CFU/ml. Based on these findings, it was concluded that there was essentially no biological activity in the high PCB-level control reactor. No plate count determinations were made for the low PCB-level control reactors.

3.4 Test Program Summary

A chronological listing of key program events is presented in Table 3 as a summary of the biphenyl and PCB biodegradation laboratory study.

TABLE 3. IMPORTANT EVENTS AND MILESTONES

Date	Day of Study	Event
2/22/88	1	Started biphenyl culture reactors
2/26/88	5	Reseeded biphenyl reactors
3/1/88	9	Began increasing salinity in biphenyl reactors (2 gr/kg per day)
3/5/88	13	Completed sea salt addition
3/9/88	17	Began biphenyl growth confirmation reactors
3/15/88	23	Began biphenyl second growth confirmation reactors
3/22/88	30	Began PCB culture development reactors
3/23/88	31	Seeded PCB culture reactors
3/25/88	33	Seeded PCB culture reactors
3/28/88	36	Seeded PCB culture reactors
3/30/88	38	Seeded PCB culture reactors
4/1/88	40	Seeded PCB culture reactors
4/17/88	56	Contents of one low level reactor submitted for sterilization testing
5/2/88	71	Began collecting presumptive composite sample from high level reactors
5/3/88	72	New set of low level PCB culture development reactors started
5/4/88	73	Seeded low level PCB culture reactors
5/6/88	75	Seeded low level reactors
5/9/88	78	Seeded low level reactors
5/11/88	80	Seeded low level reactors
5/12/88	81	Completed high level presumptive composite sample
5/12/88	81	Began high level PCB confirmation reactors
5/13/88	82	Seeded low level reactors
5/13/88	82	Seeded high level confirmation reactors
5/17/88	86	Reseeded high level confirmation reactors
5/17/88	86	Sampled high level confirmation reactors (T=0)
5/31/88	100	Sampled high level confirmation reactors (T=14)
6/14/88	114	Began collecting presumptive composite sample from low level reactors
6/23/88	123	Completed low level presumptive composite sample
6/23/88	123	Began low level PCB confirmation reactors
6/24/88	124	Seeded low level confirmation reactors
6/28/88	128	Reseeded low level confirmation reactors
6/28/88	128	Sampled low level confirmation reactors (T=0)
7/12/88	142	Sampled low level confirmation reactors (T=14)

4.0 TEST RESULTS

The raw data collected during the pilot study are included in Appendices D, E, and F. In Appendix D, the day-to-day results describing culture conditions (uptake, solids, turbidity, etc.) are presented in a spread sheet format. The test results for PCB analyses performed by GSRI and Radian are reported in Appendices E and F, respectively. The GSRI PCB data were reviewed and validated by Jordan. Jordan prepared summary tables that were submitted to Radian. These tables appear in Appendix E; analytical QA/QC data were not included in the summaries provided by E.C. Jordan. For the Radian PCB results, the summary data sheets and QA/QC data obtained from the laboratory are presented in Appendix F and G, respectively.

4.1 Biphenyl Culture Development

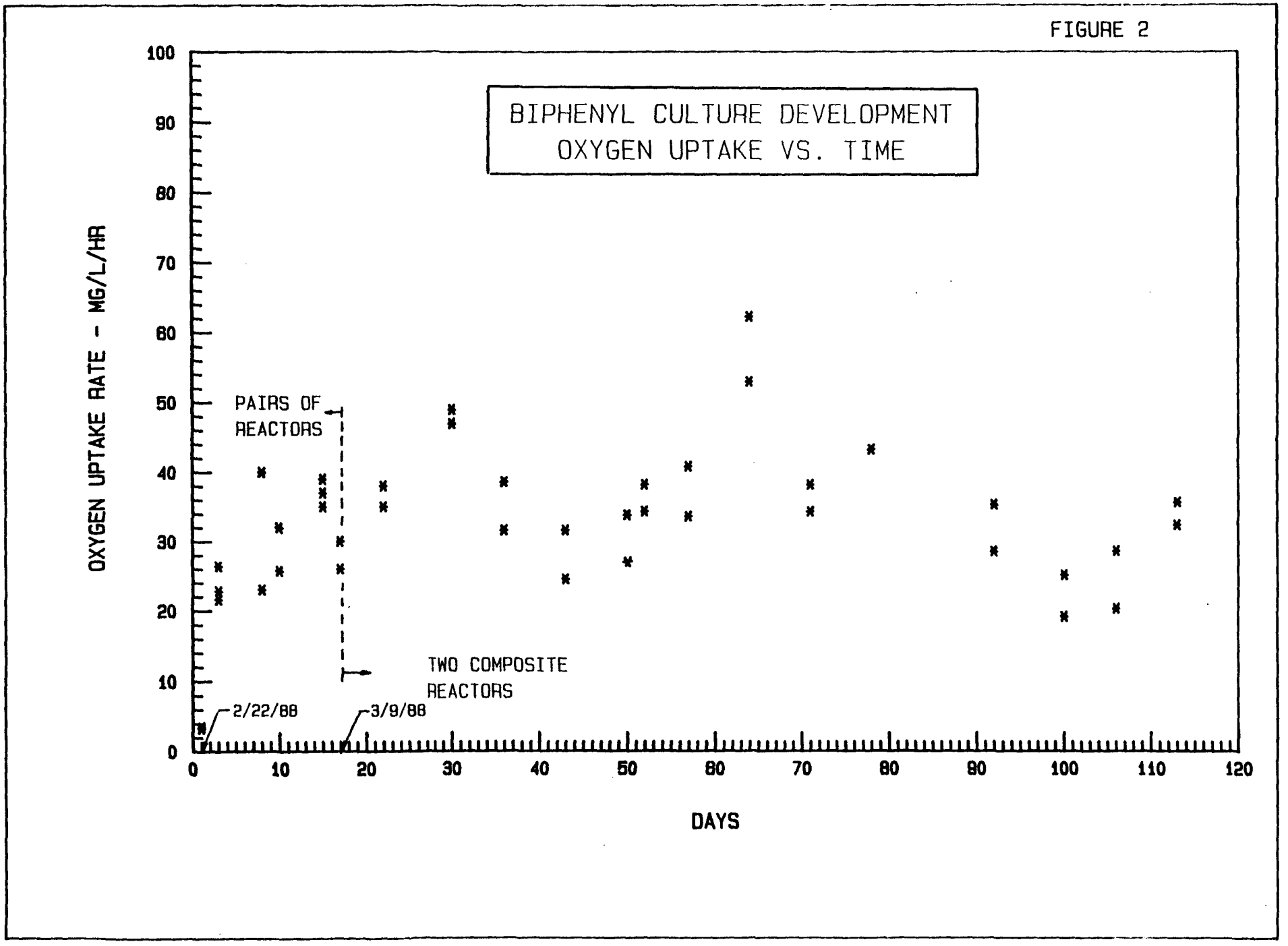
4.1.1 Acclimation

Data on the oxygen uptake rate, total solids, and total volatile solids from the initial reactors established to develop a biphenyl degrading culture are presented in Figures 2 through 4.

In Figure 2, the oxygen uptake rate is plotted versus time. For the first 17 days of the study, the uptake rate was determined on pairs of reactors selected at random. On the 17th day (March 9), two reactors were discarded, and the other four were combined into two separate flasks and maintained for the remainder of the program. These cultures were maintained primarily for seeding purposes.

The reactors experienced a very rapid increase in oxygen uptake rate. This was not unexpected based on the use of the raw sewage as the substrate. Between Days 8 and 15, the rates remained relatively stable; the increase in salinity may have contributed to this leveling off effect. The oxygen uptake rates decreased on Day 17; however, they quickly recovered and fluctuated around 40 mg/l/hr for the remainder of the study. The oxygen uptake trends shown in Figure 2 can, in most cases, be explained by the effects of sampling (solids analysis) and the use of the reactor contents for

FIGURE 2


BIPHENYL CULTURE DEVELOPMENT
OXYGEN UPTAKE VS. TIME
OXYGEN UPTAKE RATE - MG/L/HR
DAYS
PAIRS OF REACTORS
TWO COMPOSITE REACTORS
2/22/88
3/9/88
0
10
20
30
40
50
60
70
80
90
100
110
120

seed purposes. Both of these activities required removal of solids from the reactors and resulted in a temporary decrease in oxygen uptake rate.

Solids data is plotted in Figures 3 and 4. Both the total solids and total volatile solids follow the same general trend. A very rapid increase (Day 1 to 10) is followed by a plateau (Day 10 to 50). The total solids increase resulted from the added salinity and culture growth while the volatile solids increase resulted primarily from growth of the culture. Neglecting biological decay, the two means of removing volatile solids from the reactors were sampling and removal of biomass for seeding other reactors. Since the sampling schedule remained essentially the same, this should not have had a cumulative effect on the solids level. Seed material was taken from each reactor on Days 31, 33, 36, 38, and 40 (35 ml each time). This probably caused the drop in volatile solids that occurred between Day 30 and 45. The volatile solids then began to increase, reaching a new peak on about Day 79. Media was again taken for seed purposes on Days 73, 75, 78, 80 and 82. Again, the volatile solids dropped for a short period, then increased as the end of the study approached.

4.1.2 Growth Confirmation

The turbidity data developed during the biphenyl growth confirmation phase are presented in Figures 5 through 8. All test reactors showed an increase in turbidity over a relatively short period of time. The turbidity usually increased very slowly, then reached a point at which a very significant increase occurred. In general, the rate of increase was greater in the first set of growth reactors than in the second set of growth reactors. This was probably attibutable to the seeding procedure. The first growth reactors were seeded from the acclimation reactors, while the second growth reactors were seeded from the first. It can be assumed that the number of active organisms transferred from the acclimation reactors was greater than that collected from the first growth reactors. In addition, substrate carryover with the seed material transferred from the acclimation reactor to the first

FIGURE 3

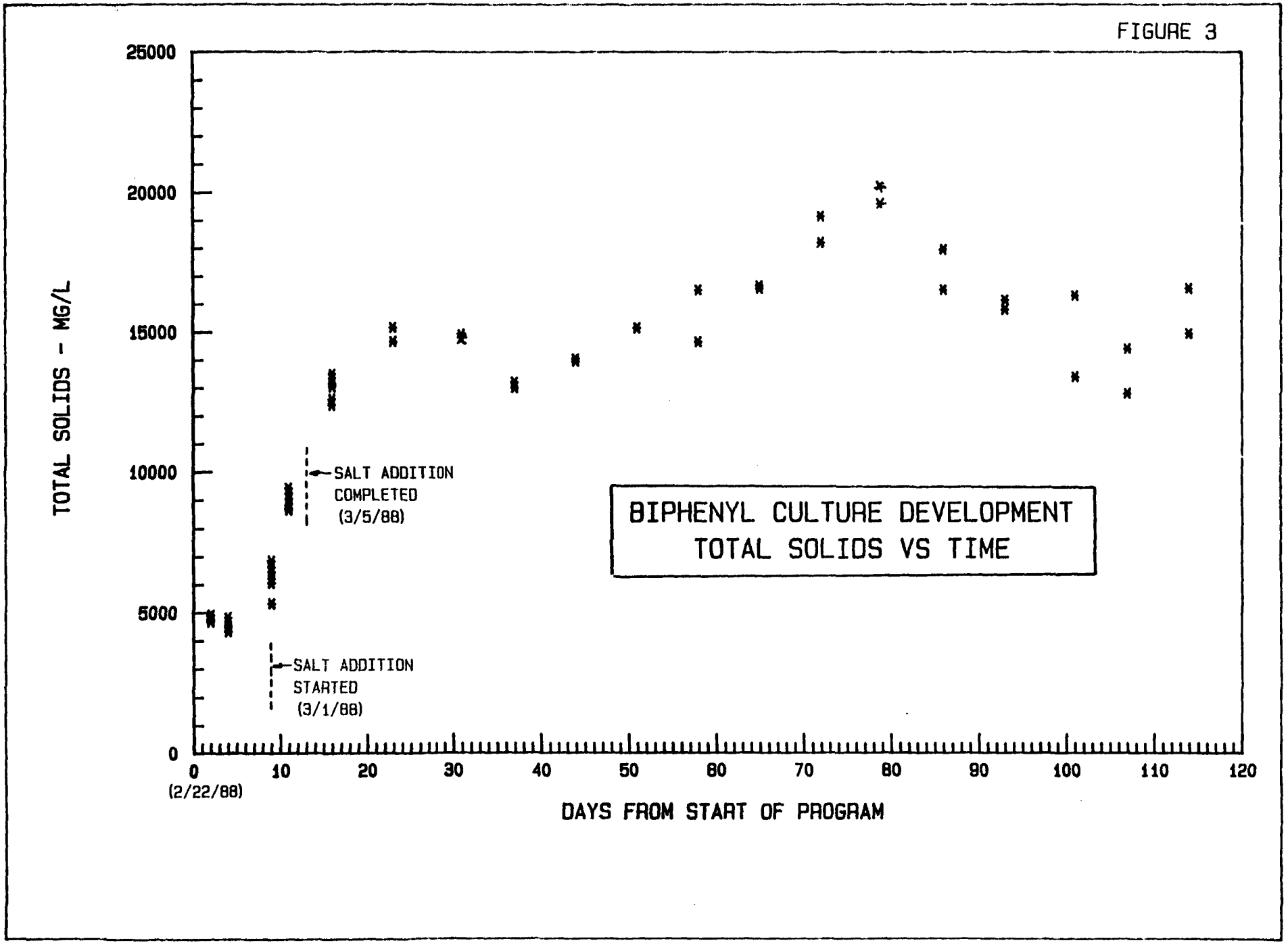

BIPHENYL CULTURE DEVELOPMENT
TOTAL SOLIDS VS TIME
TOTAL SOLIDS - MG/L
DAYS FROM START OF PROGRAM
SALT ADDITION STARTED (3/1/88)
SALT ADDITION COMPLETED (3/5/88)
(2/22/88)
25000
20000
15000
10000
5000
0
0
10
20
30
40
50
60
70
80
90
100
110
120

FIGURE 4

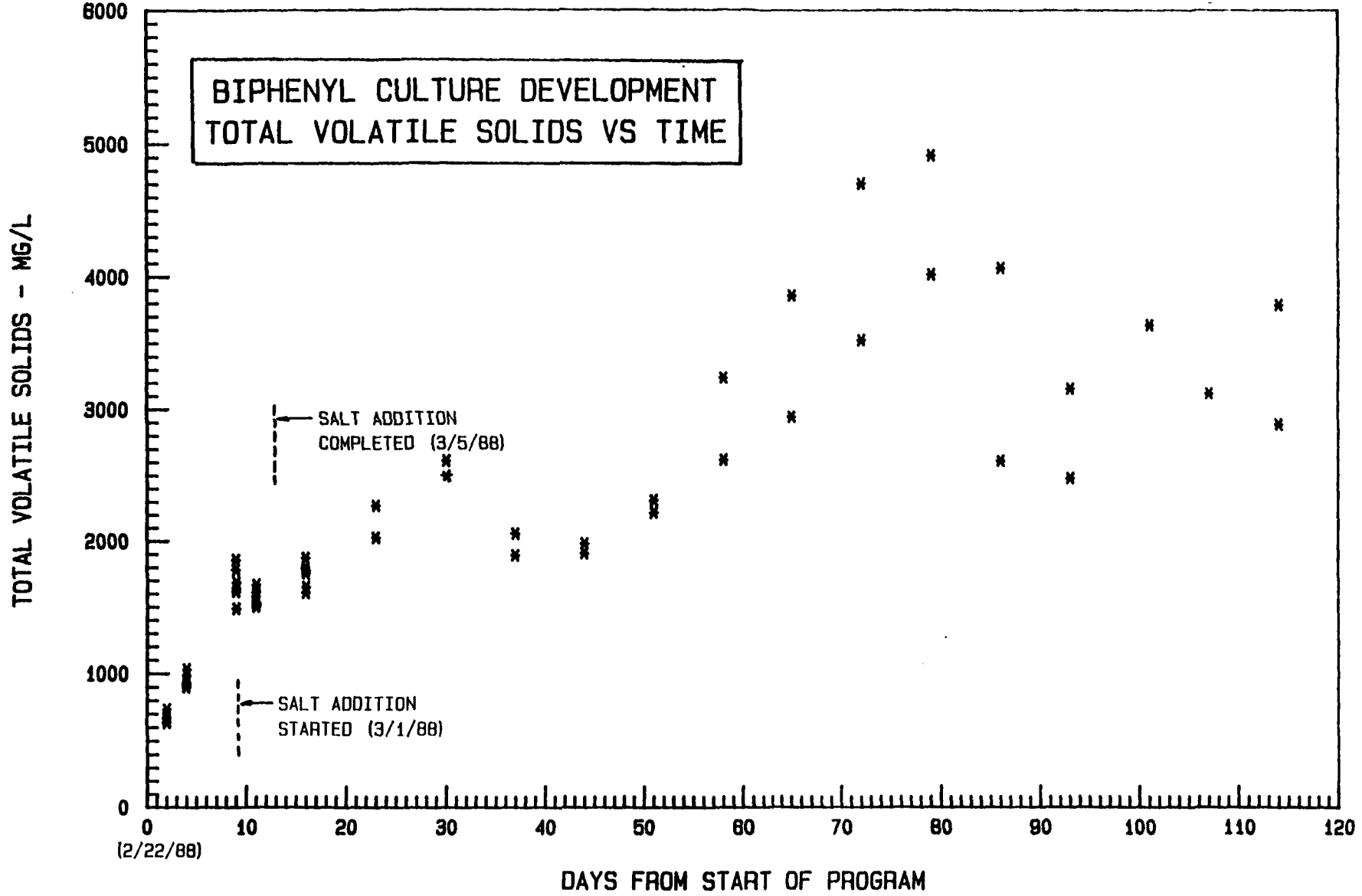

BIPHENYL CULTURE DEVELOPMENT
TOTAL VOLATILE SOLIDS VS TIME
TOTAL VOLATILE SOLIDS - MG/L
6000
5000
4000
3000
2000
1000
0
SALT ADDITION
COMPLETED (3/5/88)
SALT ADDITION
STARTED (3/1/88)
0
(2/22/88)
10
20
30
40
50
60
70
80
90
100
110
120
DAYS FROM START OF PROGRAM

FIGURE 5

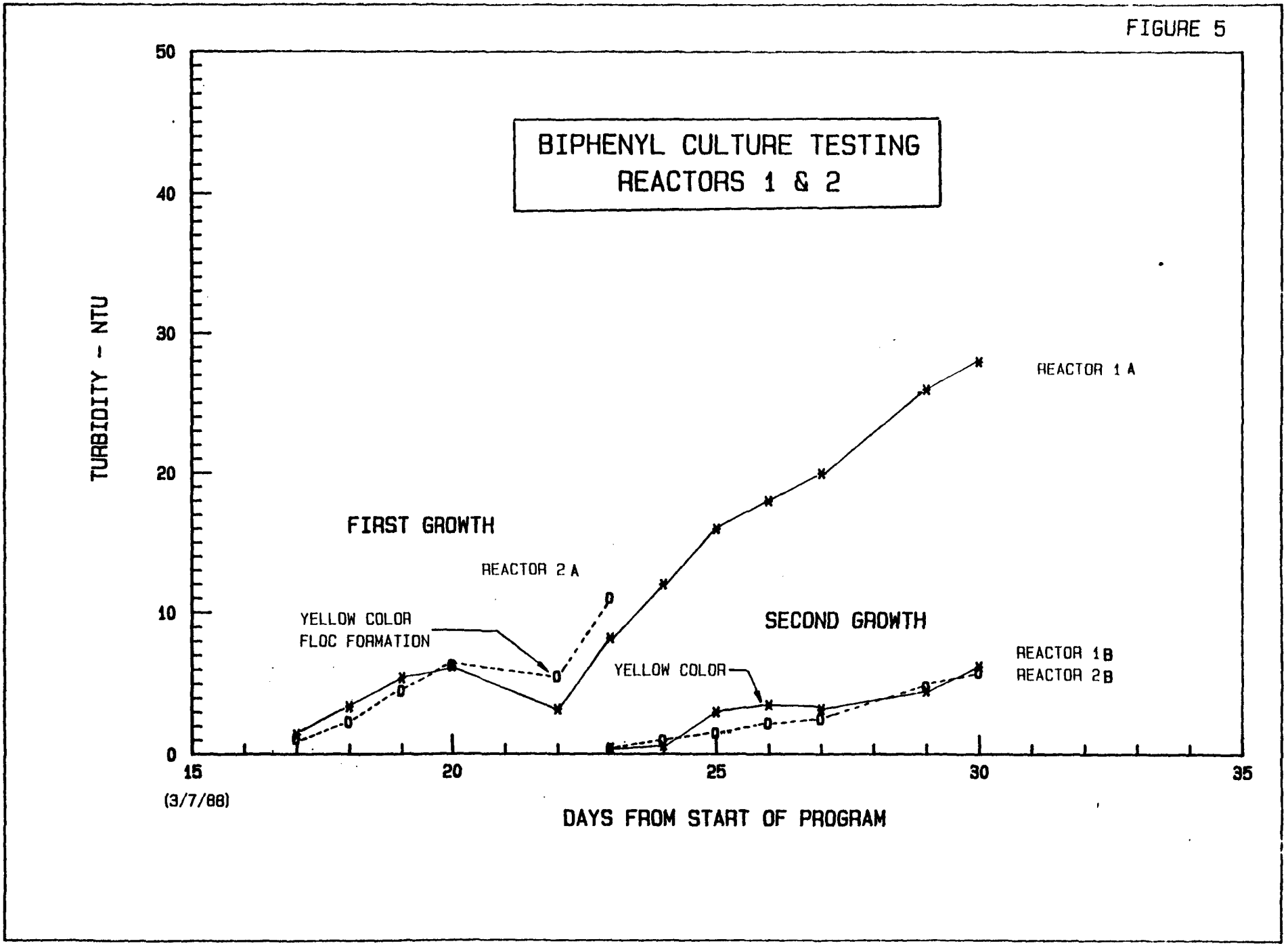

BIPHENYL CULTURE TESTING
REACTORS 1 & 2
TURBIDITY - NTU
50
40
30
20
10
0
15
20
25
30
35
(3/7/88)
DAYS FROM START OF PROGRAM
REACTOR 1 A
REACTOR 2 A
FIRST GROWTH
YELLOW COLOR
FLOC FORMATION
SECOND GROWTH
YELLOW COLOR
REACTOR 1B
REACTOR 2B

FIGURE 6

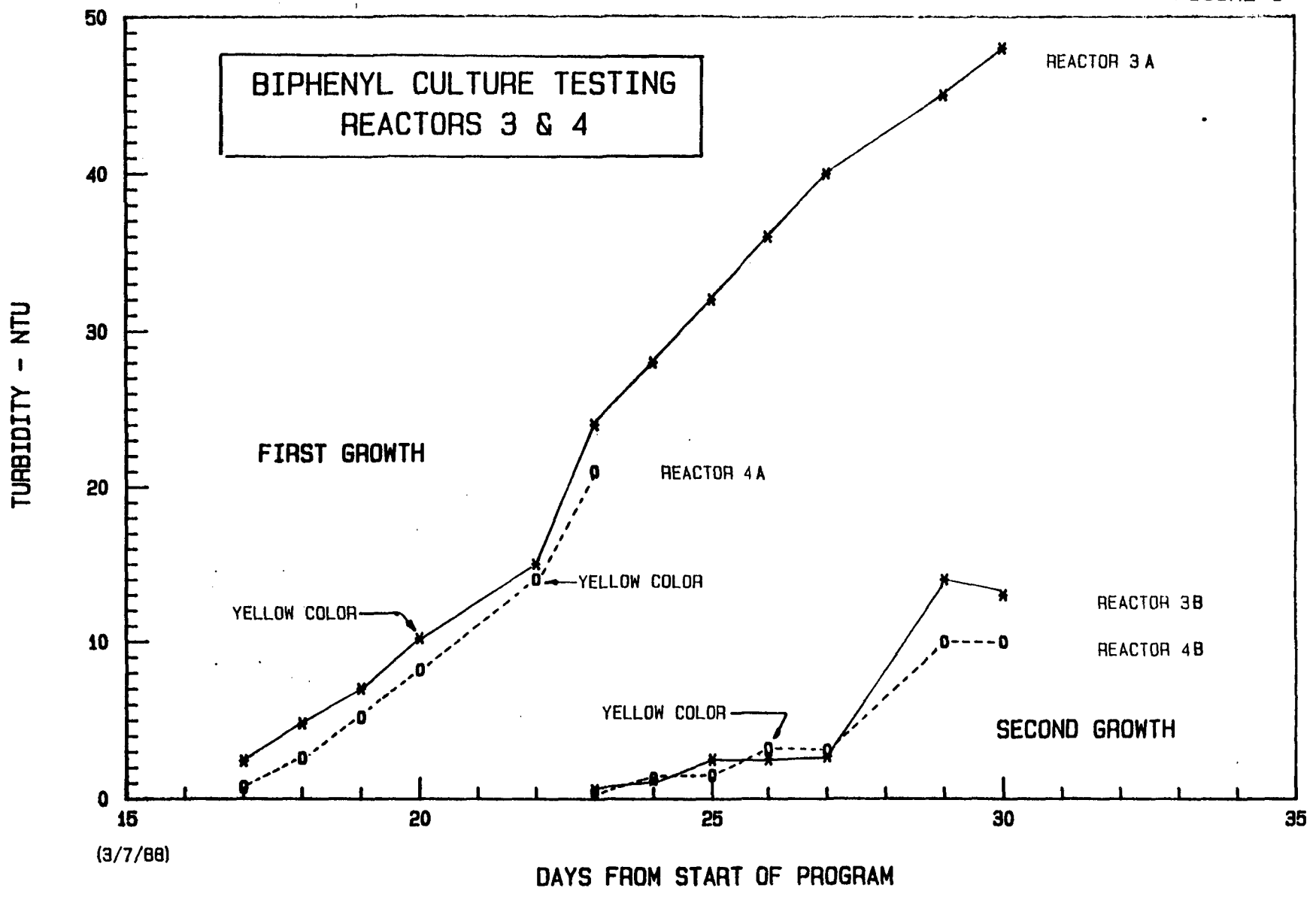

BIPHENYL CULTURE TESTING
REACTORS 3 & 4
REACTOR 3 A
REACTOR 4 A
REACTOR 3B
REACTOR 4B
FIRST GROWTH
SECOND GROWTH
YELLOW COLOR
YELLOW COLOR
YELLOW COLOR
TURBIDITY - NTU
DAYS FROM START OF PROGRAM
(3/7/88)
0
10
20
30
40
50
15
20
25
30
35

FIGURE 7

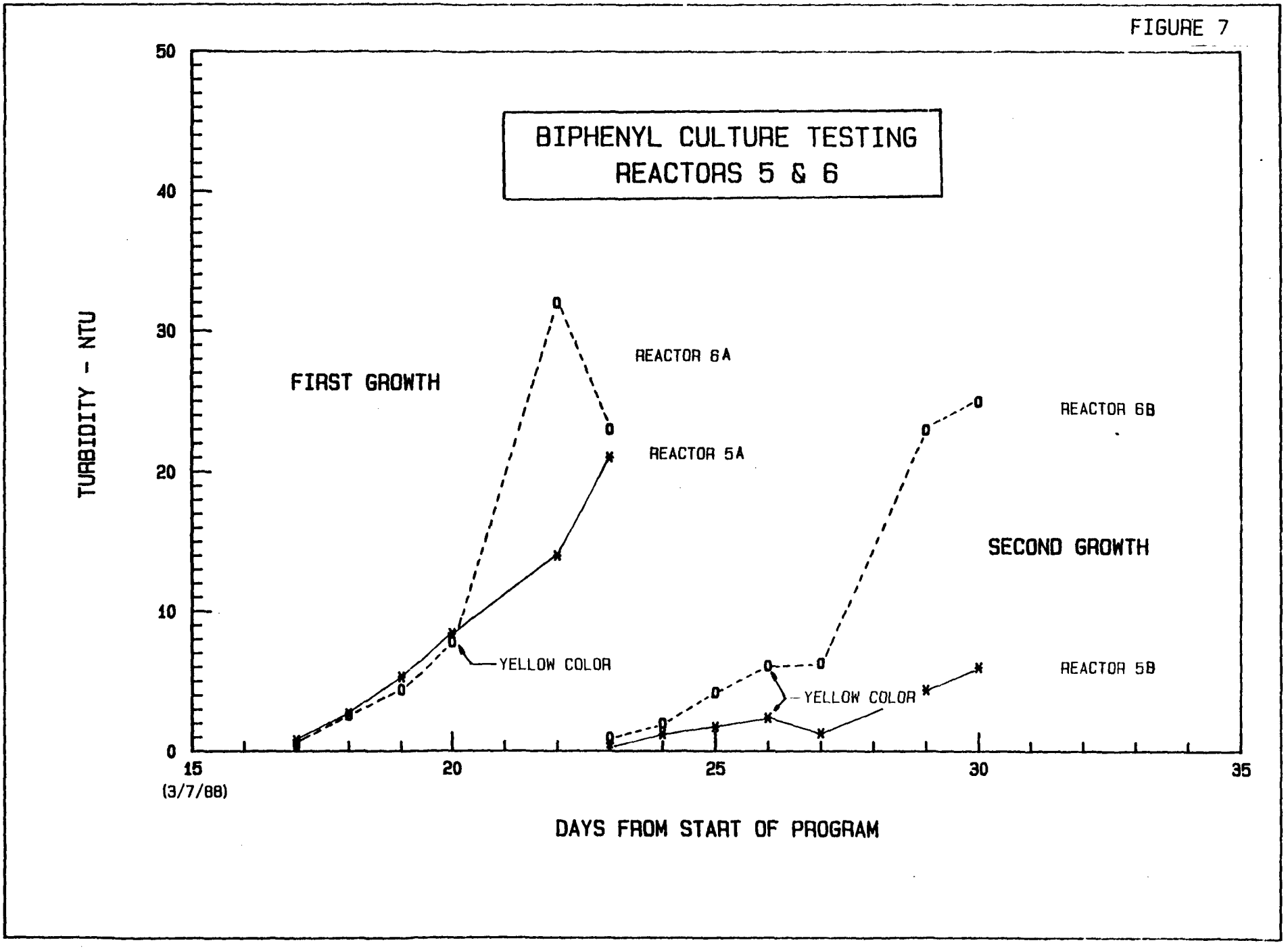
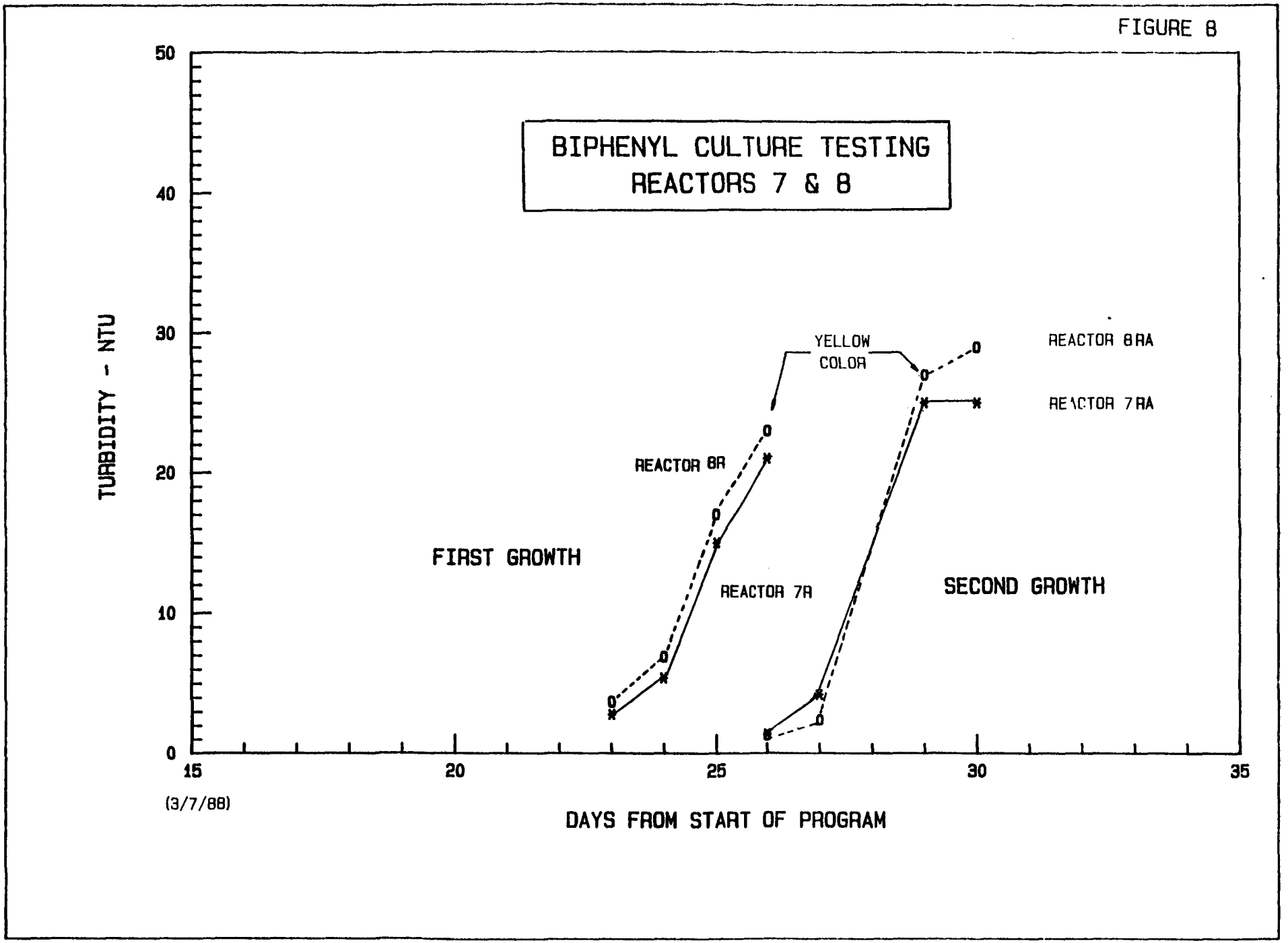

BIPHENYL CULTURE TESTING
REACTORS 5 & 6
TURBIDITY - NTU
50
40
30
20
10
0
15
(3/7/88)
20
25
30
35
DAYS FROM START OF PROGRAM
FIRST GROWTH
SECOND GROWTH
REACTOR 6A
REACTOR 5A
REACTOR 6B
REACTOR 5B
YELLOW COLOR
YELLOW COLOR

FIGURE 8


BIPHENYL CULTURE TESTING
REACTORS 7 & 8
TURBIDITY - NTU
50
40
30
20
10
0
15
20
25
30
35
(3/7/88)
DAYS FROM START OF PROGRAM
YELLOW
COLOR
REACTOR 8RA
REACTOR 7RA
REACTOR 8R
REACTOR 7R
FIRST GROWTH
SECOND GROWTH

reactor probably included the more easily metabolized sewage organics; very little if any sewage was transferred in seeding the second set of growth reactors from the first.

During this portion of the study, an additional flask was set up as a blank. The blank contained media (deionized water, sea salts, and micronutrients) and biphenyl, but no seed material. The flask was connected in series with the other six flasks by the air tubing and placed on the Burrell shaker. The blank had an initial turbidity of 0.75 NTU and remained at that level for three days. After that, the turbidity began to increase reaching 5.0 NTU on day five (see Appendix D). The original blank was discarded and a second prepared. The second blank also experienced an increase going from an initial value of 0.7 to 5.0 NTU two days later. Although no seed was added, it is believed that microorganisms capable of degrading biphenyls were introduced into the reactor by condensation in the interconnecting air tubing. Because of the apparent contamination of the blank reactors, the turbidity values plotted in Figures 5 through 8 have not been corrected for the blank values.

A color change from clear to yellow was observed in all of the biphenyl acclimation reactors (with the exception of the blank). Published studies have demonstrated that one of the degradation products of biphenyl produces a yellow color. This color change and the observed increase in turbidity were indications that a successful biphenyl-degrading culture had been developed.

4.1.3 Microscopic Examination

During each phase of the biphenyl culture development, samples were obtained and observed under a microscope. A brief summary of the results is presented below.

March 9, 1988: Sampled acclimation reactors 2, 6 and 7. Observed swimming organisms, stalked ciliates, and some nocardia.

March 15, 1988: Acclimation culture contained numerous nocardia and a few flagellates and stalked ciliates. There was a dense biological floc with some reddish colonies, possibly nitrifiers. First growth reactors contained some bacterial masses and numerous flagellates and one daphnae were observed.

March 21, 1988: Sampled second growth reactors. Observed numerous nocardia colonies and flagellates. Also some dispersed and filamentous bacteria were noted.

4.2 PCB Culture Development

4.2.1 PCB Culture Acclimation

In this phase, two PCB cultures were acclimated; low PCB concentration harbor sediment was added to one set of reactors and high PCB-level sediment was added to another set. The oxygen uptake rate is plotted versus time for the high and low PCB-level acclimation reactors in Figures 9 and 10, respectively. In the high PCB-level sediment reactor, the initial uptake rate was approximately 1.0 mg/l/hr. The seeding procedure (Days 31, 33, 36, 38, and 40) produced a marked increase in uptake (7-12 mg/l/hr), followed by a decrease when seed addition was discontinued. For the majority of the acclimation period (Days 50 to 75), the oxygen uptake rate remained relatively constant in the 7-10 mg/l/hr range. This is significantly lower than the 40 mg/l/hr uptake maintained in the biphenyl acclimation culture. Considering the expected lower biodegradability of the bottom sediments, the lower uptake is not surprising. The last sample that was taken (Day 79) showed an increase in the uptake rate; however, since additional data were not collected, it is uncertain if this increase represents an actual upward trend in oxygen uptake.

Oxygen uptake in the low PCB-level sediment reactors followed a somewhat similar trend. When the reactors were being seeded, the uptake rate increased to the 12-15 mg/l/hr range. The rate then dropped and remained in

FIGURE 9

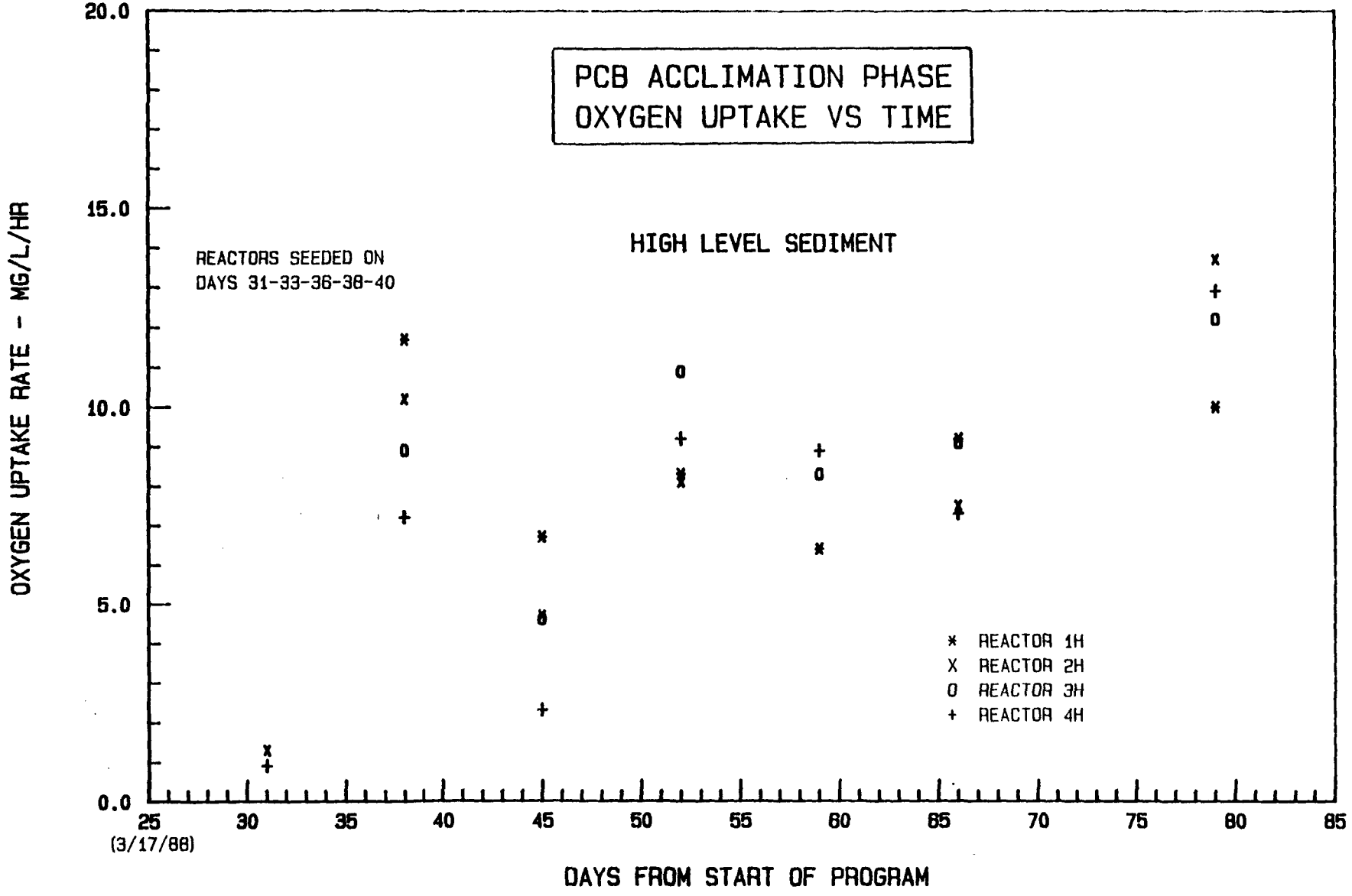
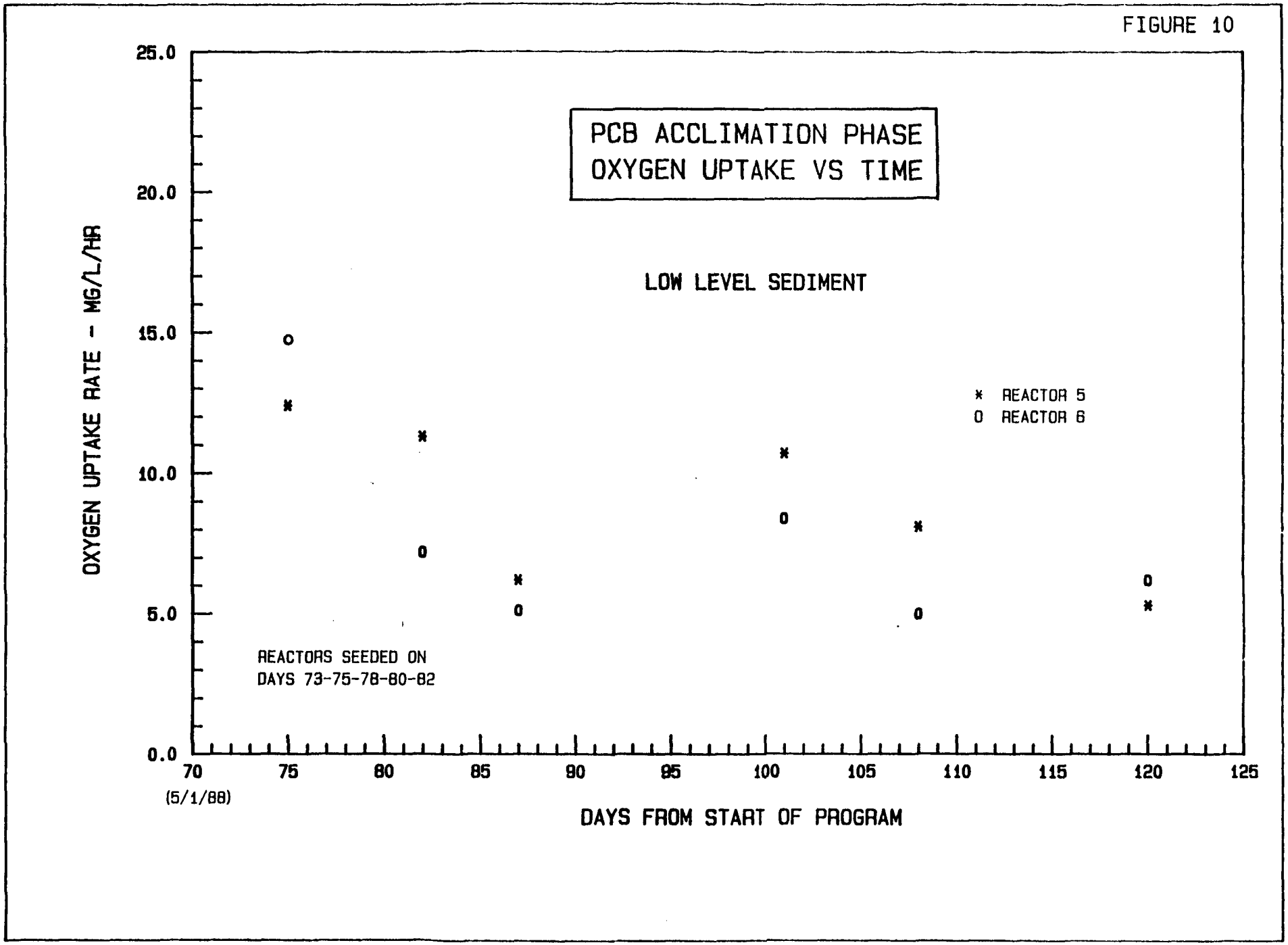

PCB ACCLIMATION PHASE
OXYGEN UPTAKE VS TIME
HIGH LEVEL SEDIMENT
REACTORS SEEDED ON
DAYS 31-33-36-38-40
OXYGEN UPTAKE RATE - MG/L/HR
20.0
15.0
10.0
5.0
0.0
25
30
35
40
45
50
55
60
65
70
75
80
85
(3/17/88)
DAYS FROM START OF PROGRAM
* REACTOR 1H
X REACTOR 2H
O REACTOR 3H
+ REACTOR 4H

FIGURE 10


PCB ACCLIMATION PHASE
OXYGEN UPTAKE VS TIME
LOW LEVEL SEDIMENT
* REACTOR 5
O REACTOR 6
REACTORS SEEDED ON
DAYS 73-75-78-80-82
OXYGEN UPTAKE RATE - MG/L/HR
DAYS FROM START OF PROGRAM
(5/1/88)
25.0
20.0
15.0
10.0
5.0
0.0
70
75
80
85
90
95
100
105
110
115
120
125

the range of 5-9 mg/l/hr for the remainder of the period. This steady state uptake rate was lower than that maintained in the high PCB-level reactors and may have been caused by the slightly different nature or composition of the sediments.

Solids data for both sets of PCB acclimation reactors (high and low PCB-level sediment) are plotted in Figures 11 through 14. For the high level sediment (Figure 11 and 12), both the total and total volatile solids concentrations remained about the same during the entire acclimation period. Although it appears that there may have been a slight upwards trend in solids concentration near the end of the program, the data do not indicate a significant increase in solids.

For the low PCB-level sediment reactors, solids concentrations actually decreased slightly over time. This was probably not an indication of reactions in the flask, but rather a function of the media added on a daily basis. On Day 89, when solids decreases were first observed, a new batch of low PCB-level sediment media was added to the reactors. Both the total and total volatile solids concentrations were lower in the new batch than in the initial batch of sediment used. When this material was used, the solids concentrations in the flasks tended to decrease to the level of the feed. On Day 102, a third batch with a higher solids concentration was prepared. As shown in Figures 13 and 14, the downward trend was reversed and the solids concentration began to increase. In general, it was concluded that very little growth or degradation of volatile solids occurred during acclimation of either the high or low PCB-level sediment reactors.

4.2.2 Presumptive Test Data

During the PCB-degradation presumptive tests, 10-day composite samples of the feed and effluent from the acclimation reactors were collected. The composites were analyzed for total solids, total volatile solids, and PCBs by Methods 608 and 680. As discussed, GSRI conducted PCB analyses on all samples while Radian analyzed for PCBs by Method 680 on select samples. The PCB lab results were submitted by GSRI to E.C. Jordan for review. Jordan validated the data and prepared a summary deleting

FIGURE 11

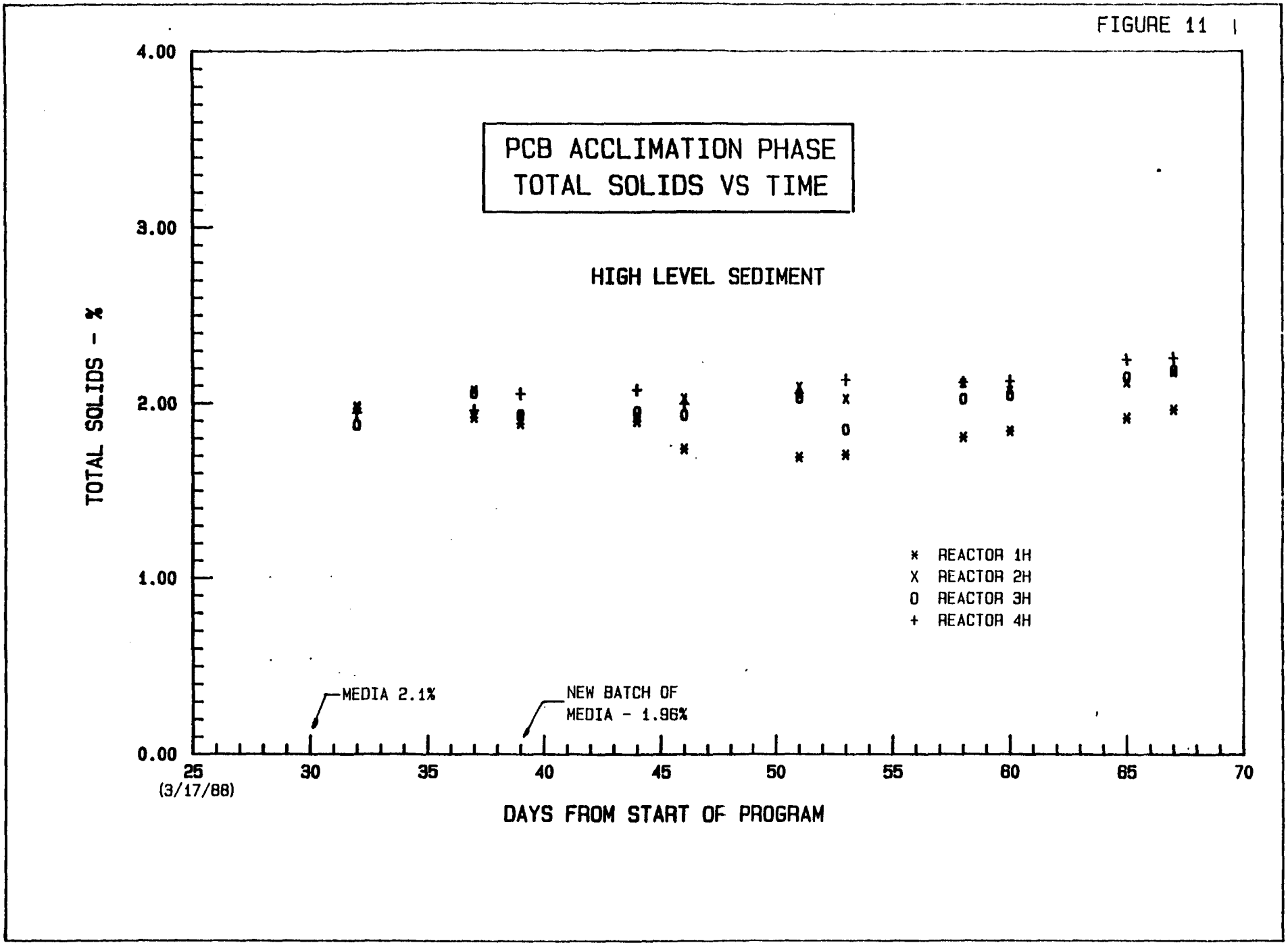

PCB ACCLIMATION PHASE
TOTAL SOLIDS VS TIME
HIGH LEVEL SEDIMENT
TOTAL SOLIDS - %
4.00
3.00
2.00
1.00
0.00
25
(3/17/88)
30
35
40
45
50
55
60
65
70
DAYS FROM START OF PROGRAM
* REACTOR 1H
X REACTOR 2H
0 REACTOR 3H
+ REACTOR 4H
MEDIA 2.1%
NEW BATCH OF
MEDIA - 1.96%

FIGURE 12

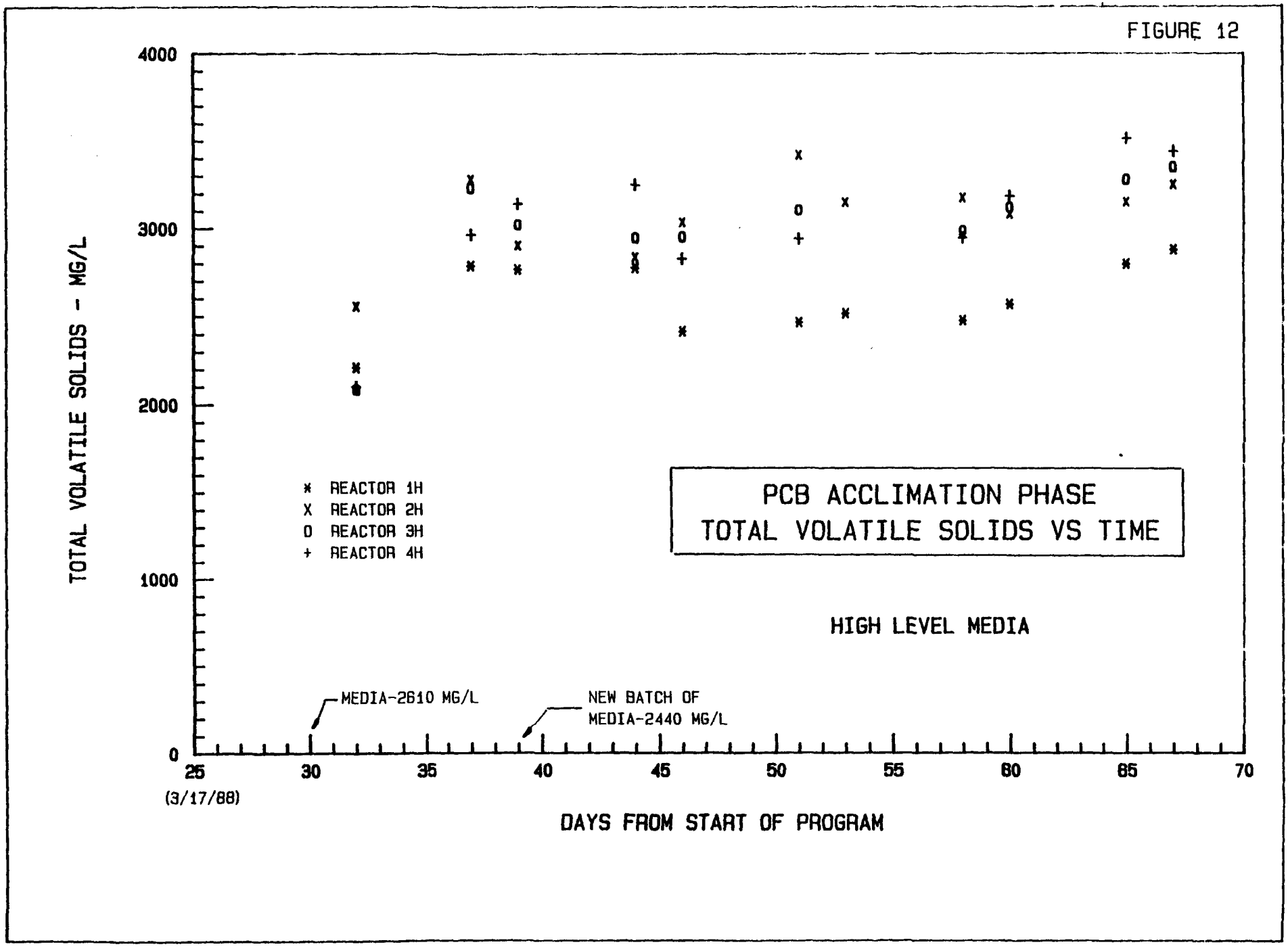

TOTAL VOLATILE SOLIDS - MG/L
4000
3000
2000
1000
0
25
30
35
40
45
50
55
60
65
70
(3/17/88)
DAYS FROM START OF PROGRAM
* REACTOR 1H
X REACTOR 2H
0 REACTOR 3H
+ REACTOR 4H
PCB ACCLIMATION PHASE
TOTAL VOLATILE SOLIDS VS TIME
HIGH LEVEL MEDIA
MEDIA-2610 MG/L
NEW BATCH OF
MEDIA-2440 MG/L

FIGURE 13

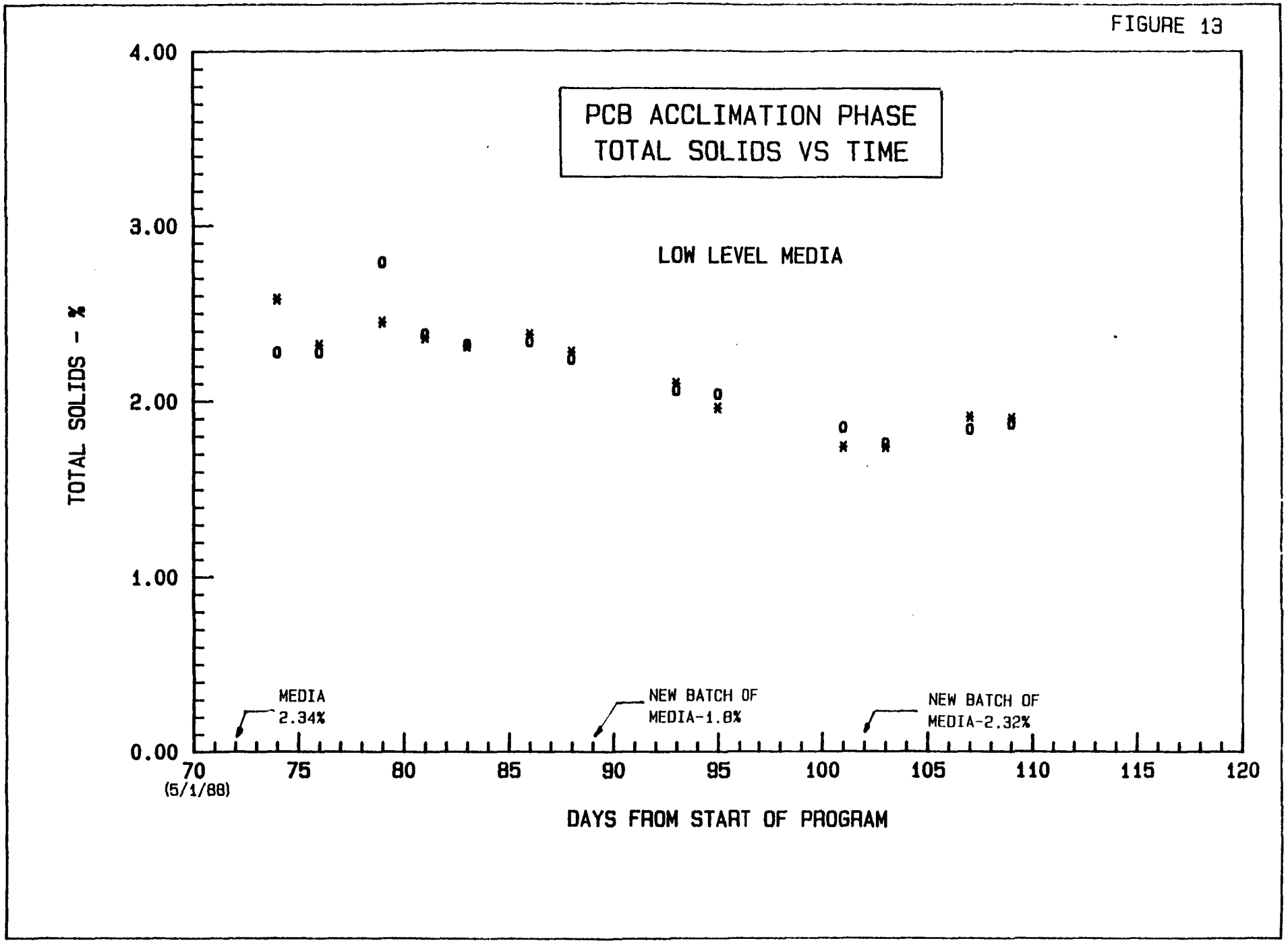
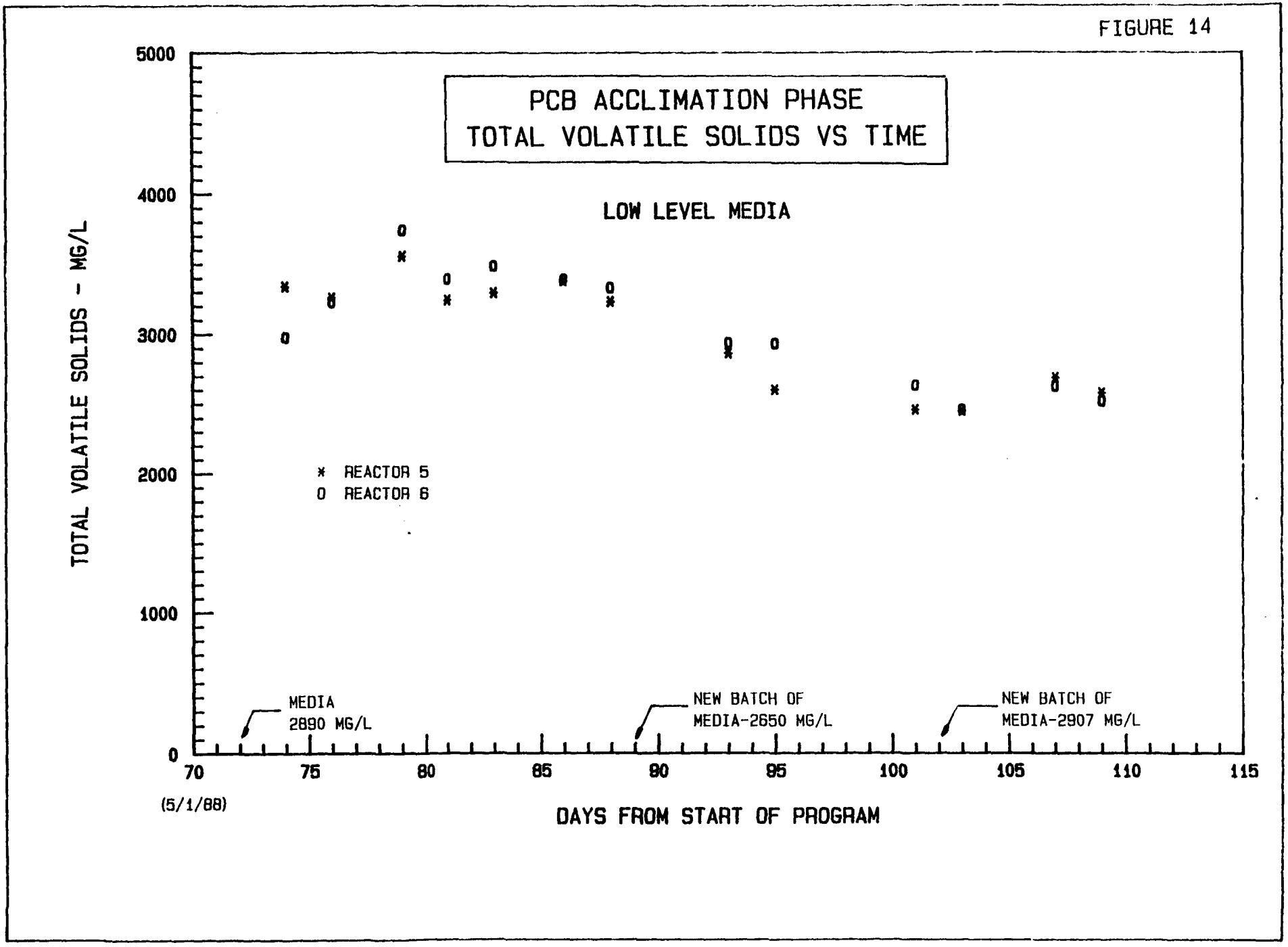

PCB ACCLIMATION PHASE
TOTAL SOLIDS VS TIME
LOW LEVEL MEDIA
TOTAL SOLIDS - %
4.00
3.00
2.00
1.00
0.00
MEDIA
2.34%
NEW BATCH OF
MEDIA-1.8%
NEW BATCH OF
MEDIA-2.32%
70
(5/1/88)
75
80
85
90
95
100
105
110
115
120
DAYS FROM START OF PROGRAM

FIGURE 14


PCB ACCLIMATION PHASE
TOTAL VOLATILE SOLIDS VS TIME
LOW LEVEL MEDIA
TOTAL VOLATILE SOLIDS - MG/L
5000
4000
3000
2000
1000
0
* REACTOR 5
0 REACTOR 6
MEDIA
2890 MG/L
NEW BATCH OF
MEDIA-2650 MG/L
NEW BATCH OF
MEDIA-2907 MG/L
70
75
80
85
90
95
100
105
110
115
(5/1/88)
DAYS FROM START OF PROGRAM

any rejected data and those values below the detection limits. The results presented in this report are based on the data prepared by E. C. Jordan. Since the GSRI data is the only complete set, it was used as the basis for evaluating the PCB-degrading performance of the reactors.

PCB analyses from the presumptive test data for the high PCB-level and the low PCB-level sediment reactors are presented in Tables 4 and 5, respectively. Results of the PCB analyses performed on five sources of sediment used to seed the reactors are presented in Table 6.

In the high PCB-level reactors, total PCB concentration decreased from 100 ppm in the influent to 85-87 ppm in three reactor effluents and to 94 ppm in one reactor effluent (P1H). The observed reduction in PCB concentration was found to correlate with an increase in total volatile solids (i.e., the lower effluent PCB levels corresponded with the highest reactor volatile solids levels). This is illustrated in Figure 15. Since increases in volatile solids typically indicate microbial growth, it appears that biological degradation of PCB occurred in the reactors. However, it should be noted that this correlation between decreasing PCB level and increasing volatile solids level was observed only in the presumptive test high PCB-level sediment reactors. It should also be noted that the decreases in PCB concentration were relatively small (e.g., 100 ppm to 85-87 ppm). Because of the accuracy of PCB analytical methods (e.g., acceptable recoveries between 50 and 150%), these decreases may not be significant.

Both Tables 4 and 5 indicate that a significant reduction in the levels of "di" and "tri" chlorobiphenyls was observed in the presumptive PCB-degradation tests. The fractional removals of these isomers were somewhat higher in the low PCB-level sediment reators. Essentially no removal of the more highly chlorinated species (i.e. tetra and penta) occurred in the high PCB-level sediment reactors and only a small percent removal of these isomers (6-14%) was observed in the two low PCB-level sediment reactors. Overall isomer removal results from the two sets of presumptive test reactors are shown in Table 7.

TABLE 4. PRESUMPTIVE TEST PHASE - PCB CONCENTRATIONS IN HIGH LEVEL SEDIMENT REACTOR INFLUENT AND EFFLUENTS

	Influent	Effluent			
PCB	AK365/366	P1H AK371/372	P2H AK360/361	P3H AK368/369	P4H AK363/364
Aroclor 1248	110[a]	97	49	82	110
Aroclor 1254	18	20	14	20	27
TOTAL	128	117	63	102	137
Isomer Groups					
Mono[b]	-	-	-	-	-
Di	6.10	6.20	1.70	2.30	1.80
Tri	33.60	28.40	20.00	22.70	22.20
Tetra	35.00	35.20	35.60	34.20	35.60
Penta	19.80	19.30	22.70	20.20	20.20
Hexa	5.20	4.60	6.40	5.50	5.70
Hepta	0.50	0.50	0.62	0.49	0.51
Octa	-	-	-	-	-
TOTAL	100.20	94.20	87.02	85.39	86.01
Total Solids, %		2.13	2.21	2.26	2.28
Total Volatile Solids, ppm		3095	3460	3615	3560

[a]All values in mg/l

[b]Monochlorobiphenyls

TABLE 5. PRESUMPTIVE TEST PHASE - PCB CONCENTRATIONS IN LOW LEVEL SEDIMENT REACTOR INFLUENTS AND EFFLUENTS

	Influent	Effluent	
PCB	AN001	P5L AN002	P6L AN003
Aroclor 1242	6.90[a,b]	-	-
Aroclor 1248	-	10.00	8.80
Aroclor 1254	1.20	1.20	0.95
TOTAL	8.10	11.20	9.75
Isomer Groups			
Mono[c]	-	-	-
Di	0.80	0.17	0.14
Tri	3.10	1.60	1.60
Tetra	2.20	1.90	1.90
Penta	1.70	1.60	1.60
Hexa	0.39	0.40	0.45
Hepta	-	-	-
Octa	-	-	-
TOTAL	8.19	5.67	5.69

[a]All values in mg/l

[b]GSRI reported that Aroclor 1242 and 1248 are difficult to distinguish in the presence of Aroclor 1254

[c]Monochlorobiphenyls

TABLE 6. PCB CONCENTRATIONS IN MICROBIAL SEEDS USED IN PRESUMPTIVE TEST HIGH AND LOW PCB-LEVEL SEDIMENT REACTORS

PCB	Intertidal Zone AK064 mg/kg	CSO AK065 mg/kg	Low Level Aerobic AK066 mg/kg	High Level Aerobic AK068 mg/kg	U of W PCB Reactor AI024 mg/l
Aroclor 1242			1100	2000	0.022
Aroclor 1248	35.0	4.6	-	-	-
Aroclor 1254	20.0	6.8	140	1300	0.120
TOTAL	55.0	11.4	1240	3300	0.142
Isomer Groups					
Mono				-	
Di				1400	
Tri				3900	
Tetra				4600	
Penta				2500	
Hexa				620	
TOTAL				13,020	
Total Solids, %	67	66	38	30	-

FIGURE 15

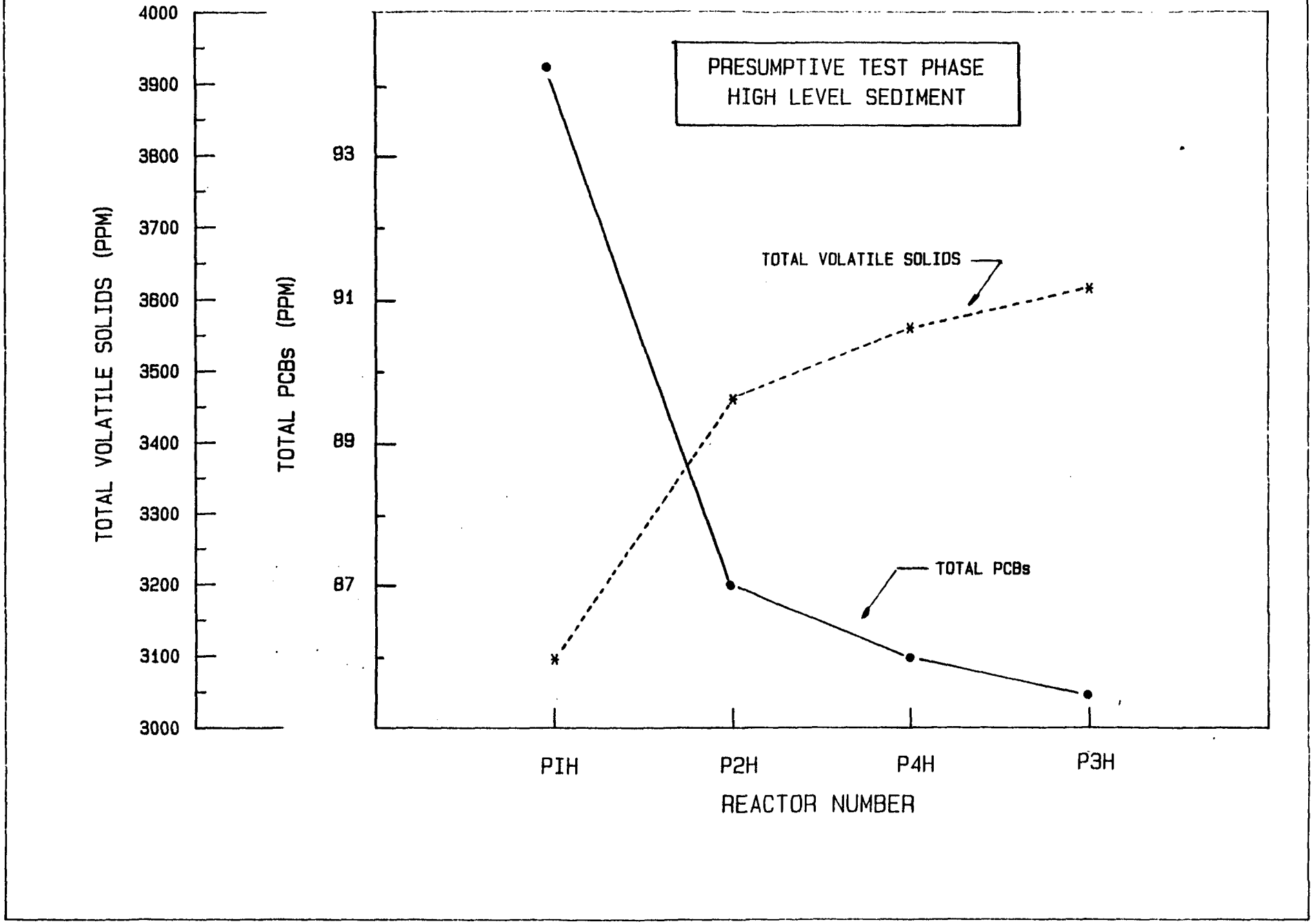

PRESUMPTIVE TEST PHASE
HIGH LEVEL SEDIMENT
TOTAL VOLATILE SOLIDS (PPM)
4000
3900
3800
3700
3600
3500
3400
3300
3200
3100
3000
TOTAL PCBs (PPM)
93
91
89
87
TOTAL VOLATILE SOLIDS
TOTAL PCBs
P1H
P2H
P4H
P3H
REACTOR NUMBER

TABLE 7. PRESUMPTIVE TEST PHASE - PERCENT REMOVAL OF PCB ISOMERS IN HIGH AND LOW PCB-LEVEL SEDIMENT REACTORS

Reactor	Di[a]	Tri	Tetra	Penta	All Groups Combined
High Level					
P1H	~0%	15%	~0%	~0%	6%
P2H	72	40	~0	~0	13
P3H	62	32	~0	~0	15
P4H	70	34	~0	~0	14
Low Level					
P5L	79	48	14	6	31
P6L	82	48	14	6	31

[a]*Dichlorobiphenyls*

A comparison of the Method 680 PCB results obtained from the GSRI and Radian laboratories is presented in Table 8. There is a wide discrepancy in the results reported by the two laboratories for PCB concentrations in samples from the high PCB-level sediment reactors. Concentrations measured by Radian were significantly lower than those reported by GSRI. Concentrations measured by the two laboratories in samples from the low PCB-level sediment reactors also differed, but to a lesser degree. Samples shipped to the two laboratories were collected at the same time using the same sampling procedure. They were also packaged and shipped in a similar fashion; therefore, the discrepancy in reported PCB concentrations appears to be a result of differences in laboratory procedures.

It is speculated that the large discrepancies between the PCB concentrations reported by the two laboratories are the result of sample homogeneity. The samples submitted to the laboratories contained both sediment and aqueous phases. Radian contacted E. C. Jordan to obtain information regarding the sample preparation being used by GSRI for PCB analysis of the sediment samples. Radian was directed to thoroughly mix the two-phase samples and then pull an aliquot for extraction. Although this procedure was used, the nonhomogeneity of the samples made it difficult to obtain a representative aliquot for extraction. Laboratory personnel indicated that pipetting from the mixed samples seemed to restrict the amount of solids in the aliquots used for PCB analysis. Since PCBs are typically associated with solids, low sediment content in the aliquots analyzed probably accounted for the low PCB concentrations reported by Radian.

Quality control data for the PCB analyses conducted by Radian are presented in Appendix G (a summary of the PCB results obtained from the Radian laboratory appear in Appendix F). Of 80 surrogate spikes analyzed in this study, only four spikes (5 percent) fell outside of the 50% to 150% recovery control limits.

Information is not available to determine if the sample preparation procedure provided to Radian was actually used by GSRI during the study. There is an alternative sample preparation procedure which involves

TABLE 8. DATA COMPARISON - GSRI AND RADIAN RESULTS

PRESUMPTIVE PHASE

HIGH LEVEL SEDIMENT

Isomer Groups	Influent GSRI	Influent RAD.	Effluent P2H GSRI	Effluent P2H RAD.	Effluent P4H GSRI	Effluent P4H RAD.
Mono[a]	-	0.003[b]	-	-	-	-
Di	6.10	0.790	1.70	0.290	1.80	0.220
Tri	33.60	3.700	20.00	5.200	22.20	4.300
Tetra	35.00	3.700	35.60	9.000	35.60	0.720
Penta	19.80	1.100	22.70	2.900	20.20	2.200
Hexa	5.20	0.620	6.40	1.700	5.70	1.200
Hepta	0.50	0.080	0.62	0.230	0.51	0.160
Octa	-	0.011	-	0.032	-	0.022
Nono	-	-	-	0.004		0.003
TOTAL	100.20	10.004	87.02	19.356	86.01	8.825

LOW LEVEL SEDIMENT

Isomer Groups	Influent GSRI	Influent RAD.	Effluent P5L GSRI	Effluent P5L RAD.	Effluent P5L RAD.	Effluent P6L GSRI	Effluent P6L RAD.
Mono	-	-	-	-	-	-	-
Di	0.80	0.430	0.17	0.120	0.150	0.14	0.069
Tri	3.10	1.800	1.60	1.200	1.400	1.60	1.000
Tetra	2.20	1.600	1.90	1.800	2.100	1.90	1.700
Penta	1.70	0.600	1.60	0.670	0.780	1.60	0.660
Hexa	0.39	0.270	0.40	0.320	0.360	0.45	0.310
Hepta	-	0.030	-	0.040	0.046	-	0.038
Octa	-	-	-	-		-	-
TOTAL	8.19	4.73	5.67	4.15	4.836	5.69	3.777

[a] Monochlorobiphenyls

[b] All values in mg/l

separation of the aqueous and sediment phases, with subsequent extraction and PCB analysis of both phases. If this method was used by GSRI, this could account for the discrepancy between concentrations measured by the two laboratories. GSRI conducted a PCB analyses on all samples collected over the course of the study; therefore, the GSRI data were used to interpret study results.

4.2.3 Confirmation Test Data

During the confirmation phase, five reactors were operated containing high PCB-level media, and three containing low PCB-level media. In each set, formaldehyde was added to one reactor to eliminate biological activity. The reactors were sampled after the second seeding (T=0) and again 14 days later (T=14). Samples were analyzed for PCB by Methods 608 and 680. All samples were analyzed by GSRI, whereas only select samples were analyzed by Radian using Method 680. Total solids and total volatile solids analysis were also performed.

Data from the high PCB-level sediment reacotrs are presented in Table 9 and data from the low PCB-level sediment reactors are presented in Table 10. The data presented in these tables were based on the analysis performed by GSRI and submitted to Radian by E. C. Jordan.

Confirmation testing results indicate that significant PCB reduction was obtained during a 14-day period of operating the high PCB-level sediment reactors. Removals of the "di" and "tri" chlorobiphenyls was in the 60-100% range in all four reactors. In addition, with the exception of reactor C2H, varying levels of removal of the higher chlorinated groups was also noted. The percent removals measured for each isomer group are summarized in Table 11. For reactor C1H, the initial sample (T=0) was analyzed in duplicate and the removal efficiency was based on the average of the two analyses.

TABLE 9. CONFIRMATION TEST PHASE - PCB CONCENTRATIONS IN HIGH LEVEL SEDIMENT REACTORS

PCB	Reactor C1H T=0 AK362/367	Reactor C1H T=0 AK362/367	Reactor C1H T=14 AK382	Reactor C2H T=0 AK374/375	Reactor C2H T=14 AK383
Aroclor 1242	-			-	
Aroclor 1248	86.00[a]			80.00	
Aroclor 1254	15.00			13.00	
	101.00			93.00	
Isomer Groups					
Mono[b]	-	-	-	-	-
Di	3.30	3.00	0.54	2.30	0.13
Tri	29.90	27.00	9.90	21.80	7.70
Tetra	35.30	32.00	25.60	27.30	25.30
Penta	20.00	17.00	13.10	14.30	13.80
Hexa	5.40	4.20	3.20	3.30	3.60
Hepta	0.46	0.14	0.19	0.20	0.20
TOTAL	94.36	83.34	52.53	69.20	50.73
Total Solids, %	2.12	-	2.04	2.14	2.19
Total Volatile Solids, mg/l	3170	-	2985	3205	3750

[a]All values in mg/l

[b]Monochlorobiphenyls

TABLE 9. (CONTINUED)

PCB	Reactor C3H T=0 AK376 377	Reactor C3H T=14 AK384	Reactor C4H T=0 AK378 379	Reactor C4H T=14 AK385	Reactor 7CCH T=0 AK380 381	Reactor 7CCH T=14 AK386
Aroclor 1242	-		-		100.00	
Aroclor 1248	100.00[a]		94.00		-	
Aroclor 1254	17.00		15.00		35.00	
TOTAL	117.00		109.00		135.00	
Isomer Groups						
Mono[b]	-		-	-	0.61	0.21
Di	2.90	0.30	2.60	-	19.90	4.10
Tri	26.00	6.50	24.40	3.10	56.90	12.00
Tetra	28.00	23.00	28.40	10.70	54.90	12.20
Penta	16.00	12.80	15.10	6.70	26.90	6.50
Hexa	4.50	3.30	3.90	1.90	6.30	1.80
Hepta	0.43	0.18	0.34	-	0.46	-
TOTAL	77.83	46.08	74.74	22.40	165.97	36.81
Total Solids, %	2.18	2.12	2.26	2.02	2.26	2.04
Total Volatile Solids, mg/l	3245	3475	3425	3120	3220	2985

[a]All values in mg/l

[b]Monochlorobiphenyls

TABLE 10. CONFIRMATION TEST PHASE - PCB CONCENTRATIONS IN LOW LEVEL SEDIMENT REACTORS

PCB	Reactor C5L T=0 AN004	Reactor C5L T=14 AK871	Reactor C6L T=0 AN005	Reactor C6L T=14 AK872	Reactor 8CCL T=0 AN006	Reactor 8CCL T=14 AK873
Aroclor 1241					7.40	7.40[a]
Aroclor 1248	12.00[b]	2.60	14.00	3.10	-	-
Aroclor 1254	1.20	2.60	1.30	3.10	0.91	2.90
TOTAL	13.20	5.20	15.30	6.20	8.31	10.30
Isomer Groups						
Mono[c]	-	-	-	-	-	-
Di	0.24	0.12	0.18	0.11	1.70	1.50
Tri	2.00	1.80	1.70	1.60	4.30	4.20
Tetra	2.20	2.50	1.70	2.50	2.80	2.70
Penta	1.90	1.80	1.50	1.90	2.10	1.90
Hexa	0.54	0.40	0.38	0.58	0.45	0.48
TOTAL	6.88	6.62	5.46	6.69	11.35	10.78
Total Solids, %	2.22	2.20	2.22	2.12	2.11	2.10
Total Volatile Solids, mg/l	2633	2898	2795	2675	3170	2144

[a]All values in mg/l

[b]GSRI reports - it is difficult to distinguish between 1241 & 1248 when 1254 is present

[c]Monochlorobiphenyls

TABLE 11. CONFIRMATION PHASE - PCB REMOVAL RATES

HIGH LEVEL MEDIA

Isomer Group	Reactor C1H	C2H	C3H	C4H	7CCH
Mono	-	-	-	-	66%
Di	83%	94%	90%	100%	79
Tri	65	64	75	87	79
Tetra	24	7	18	62	78
Penta	29	3	20	56	76
Hexa	33	0	27	51	71
Hepta	37	0	58	100	100
TOTAL	41	27	41	70	78

In this phase of the study, formaldehyde was added to the control reactors (7CCH and 8CCL) to eliminate any biological activity. Removal of PCBs in the control reactors was attributed to nonbiological mechanisms such as adsorption on the reactor walls or volatilization. In the initial sample (T=0), the reported PCB levels in both control reactors were approximately twice that of the corresponding active reactors, despite the fact that the same media was used. This indicates that the formaldehyde in some way affected the amount of PCBs released during the extraction step. Without representative control data, quantitative conclusions regarding biological PCB degradation could not be drawn. However, the data indicate that biological degradation of "di" and "tri" isomers occurred. In the high level control reactor, the 14-day removal rate was nearly uniform (70-80%) for all congeners (Table 11). This nonpreferential removal of isomers indicates physical/chemical transformations. Alternatively, in the four active reactors, there was a selective removal of "di" and "tri" isomers; this indicates that biological degradation of these isomer groups occurred in the active reactors.

The PCB removal efficiencies were not as great in the low PCB-level sediment reactors as in the high PCB-level media. Dichlorobiphenyl was reduced 40-50%, but little or no removal of any of the other isomer groups was noted. As with the high PCB-level reactors, this preferential reduction was in contrast to the control reactor where there was a relatively uniform reduction (0-12%) for all isomer groups (Table 10). These data are consistent with the high PCB-level reactor data, as well as with the presumptive test data.

During the confirmation phase, select samples were also submitted to Radian laboratories for PCB analysis by Method 680. The data comparison which is presented in Table 12, shows there was again a significant difference between the two data sets. As was discussed previously, these differences are probably the result of sample nonhomogeneity and differences in sample handling procedures.

TABLE 12. DATA COMPARISON - GSRI AND RADIAN RESULTS
CONFIRMATION PHASE

	Reactor C2H T=0		Reactor C2H T=14		Reactor C5L T=0		
Congeners	GSRI	RAD	GSRI	RAD	GSRI	RAD	RAD
Mono	-	-	-	- -	-	-	
Di	2.30	0.510	0.13	0.400	0.24	0.085	0.110
Tri	21.80	6.900	7.70	7.200	2.00	1.100	1.400
Tetra	27.30	8.300	25.30	30.000	2.20	1.900	2.200
Penta	14.30	2.400	13.80	7.300	1.90	0.730	0.860
Hexa	3.30	1.300	3.60	3.300	0.54	0.350	0.410
Hepta	0.20	0.160	0.20	0.410	-	0.042	0.049
Octa	-	0.021	-	0.054	-	0.006	0.005
Nona	-	0.003	-	0.019	-	-	-
	69.20	19.594	50.73	48.683	6.88	4.213	5.034

	Reactor C5L T=14			Reactor 7CCH T=14		Reactor 8CCL T=14	
Isomer Groups	GSRI	RAD	RAD	GSRI	RAD	GSRI	RAD
Mono	-	-	-	0.21	0.65	-	0.004
Di	0.12	0.041	0.057	4.10	11.00	1.50	0.430
Tri	1.80	0.610	0.840	12.00	45.00	4.20	1.900
Tetra	2.50	1.300	1.900	12.20	53.00	2.70	1.900
Penta	1.80	0.530	0.750	6.50	17.00	1.90	0.740
Hexa	0.40	0.250	0.360	1.80	8.20	0.48	0.360
Hepta	-	0.028	0.042	-	1.10	-	0.042
Octa	-	0.005	0.006	-	0.16	-	0.007
Nona	-	-	-	-	0.06	-	-
	6.62	2.764	3.955	36.81	136.17	10.78	5.383

All values reported as mg/l

5.0 SUMMARY

The results of this project indicate that biphenyl and PCBs were successfully biodegraded in an aerobic reactor and in a media with a salinity similar to that found in a brackish water environment such as the estuary in New Bedford Harbor. PCBs sorbed to New Bedford Harbor sediments were biodegraded to a limited extent by microbes enriched from sources in the estuary of New Bedford Harbor and from an anaerobic digester used to treat PCB-contaminated sewage sludge in Madison, Wisconsin. Di- and trichlorobiphenyls were preferentially removed from reactors under the conditions of this study, indicating that these isomers were biodegraded.

5.1 Biphenyl Culture Development

This portion of the program included three phases. In the first, laboratory-scale reactors were operated to develop a biphenyl degrading culture using domestic raw sewage as the primary carbon source. Once growth had been established, two additional sets of reactors were operated. The domestic sewage was eliminated to establish the culture's ability to grow with biphenyl as the only carbon source. The growth of two generations of microbial comunities with biphenyl as the only added carbon source provided evidence that a culture capable of degrading biphenyl in a brackish water environment was successfully developed. This growth was confirmed by an increase in turbidity and verified by microscopic examination.

5.2 PCB Degradation Presumptive Tests

The PCB degradation presumptive tests consisted of the operating laboratory-scale aerobic reactors in a daily fill and draw mode. The amount of PCB contaminated material added and removed daily resulted in an average hydraulic retention time of 14 days. After an acclimation period, there was found to be as much as 90% reduction of PCB isomers with three or fewer chlorines, but little, if any, measurable reduction of the higher chlorinated isomers. This differential reduction is consistent with prior research indicating that the lesser chlorinated PCB congeners are the most readily biodegraded.

The degree of PCB removal measured in reactors with lower PCB concentrations was twice that measured in reactors with higher PCB concentrations. The difference in the percentage of reduction was probably the result of an increase in the number of isomer groups degraded (i.e., levels of "tetra" and "penta" isomeric groups were reduced in the low level reactors, but not in the high level reactors). There was no large difference in the percent reduction of "di" and "tri" isomers observed in the two sets of tests.

5.3 PCB Degradation Confirmation Tests

The initial goal of the PCB degradation confirmation studies was to perform a PCB mass balance around batch operated reactors; this goal was not achieved because the initial PCB level in the control digester was found to be twice that in the corresponding test reactors (probably because of an effect of formaldehyde on the PCB extraction procedure; the same media was used in both the active and control reactors). Therefore, the amount of PCBs removed by biological mechanisms could not be differentiated from the amount that was removed by physical/chemical processes.

The pattern of isomer removal in these batch reactors was similar to that observed in the presumptive tests; the greatest amount of reduction was observed in the lesser chlorinated isomers.

Anomalies noted in control tests using sterilized cultures indicate that formaldehyde is not a suitable bactericide for use in estimating PCB biodegradation in sediments. It is possible that formaldehyde affected the PCB analysis by changing the characteristics of PCB sorption on the sediment.

5.4 Optimization of PCB Reduction

The presumptive test results showed an average percent PCB reduction of about 30 to 40%. These values do not necessarily represent the maximum amount of reduction that could be achieved. The overall goal of this project was to determine the feasibility of biodegrading PCBs in a brackish water

environment and the extent of degradation that would occur under conditions simulating a full-scale aerobic treatment system. The project plan was not designed to give kinetic data on PCB destruction or to optimize the rate of PCB reduction.

There are several potential mechanisms for enhancing the rate of PCB degradation. For example, increasing the desorption rate, enhancing cometabolism, and manipulating reactor operation modes and population characteristics could enhance PCB removal. None of these methods would likely achieve practical benefits for the treatment of New Bedford Harbor sediments, unless a mechanism to degrade all PCB isomer groups was developed. To do this, alternative biological process configurations would have to be considered that would utilize different degradative mechanisms to supplement or complement those mechanisms that occurred in the aerobic systems used in this study. For example, anaerobic dechlorination to convert higher chlorinated PCBs to lesser chlorinated isomers might be effective before aerobic treatment. However, a considerable amount of research would be required to assess the effectiveness of such alternative process configurations for enhancing PCB biodegradation in New Bedford Harbor sediments.

APPENDICES

APPENDIX A

REVISED TECHNICAL AND COST PROPOSALS

RADIAN
CORPORATION

REVISED TECHNICAL AND COST PROPOSALS

Bench Scale Testing of
Biodegradation Technologies for
PCBs in New Bedford (MA)
Harbor Sediment

Inquiry No. 4236-LAB-0081

Prepared for: EBASCO Services Inc.
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July 23, 1987

RADIAN
CORPORATION

DEVELOPMENT OF AN ENRICHED SALTWATER MICROBIAL COMMUNITY CAPABLE OF BIODEGRADING PCBs SORBED TO HARBOR SEDIMENTS

TECHNICAL APPROACH/WORKSCOPE

1.0 OVERVIEW

While microbial communities capable of degrading PCBs in fresh water, in sewage and in sediment and water mixes have been developed and demonstrated, a similar salt water system has not been developed or demonstrated.

The goal of this project phase is to develop a salt water microbial community capable of degrading PCBs and demonstrate this capability when the PCBs are sorbed to harbor sediments - specifically sediments from the harbor at New Bedford, MA. We will use microbial communities taken from different areas in the harbor, microbial communities from sewage treatment works, from fresh water PCB digestion cultures, and from purchased cultures as seeds in the development of the culture. Instead of PCBs we will use the addition of biphenyl, the parent compound of PCBs, as the selection mechanism for the microbial community for initial culture development. Then we will shift to actual PCB contaminated sediments for testing purposes.

2.0 TECHNICAL APPROACH

2.1 Selection Techniques

We will base our selection technique on developing a complete microbial community adapted to degrading PCBs in the presence of other substrates and other microorganisms. This is in contrast to simply isolating a microbial population capable of degrading PCBs in an isolated system, the process more often used in developing an enriched culture.

We chose the open culture procedure for three main reasons. First and most importantly, it would be prohibitively expensive to prevent the introduction of alien microorganisms in a full scale treatment unit. Thus it is important that the community developed must be able to compete with other microorganisms. Second, since there are numerous substrates which can serve as energy sources in the bay sediments, it is important that the community selected be able to degrade PCBs in the presence of other substrates. And third, because many authors have suggested that microorganisms degrading PCBs are doing so cometabolically, that is, using other substrates as the energy source, it may be important to have alternative energy sources available (particularly those which are analogs to PCBs, and would require a similar enzyme system for degradation).

If we attempted to develop an enriched culture by simply isolating PCB degrading microorganisms, we would not know if the developed culture would be capable of surviving and biodegrading PCBs in the full scale treatment system environment.

We will develop the culture from seven sources of microorganisms, each of which may contain microorganisms capable of degrading PCBs. We will use the idea, as has been found in developing communities for treating other recalcitrant compounds, that nature has already performed a great deal of selection and sorting. That is, if one wants to find a PCB degrading microorganism capable of doing so in a certain environment, it is best to look in that environment or a similar one where the PCBs are and have been present for a length of time.

We propose to use four sources of microbes from the New Bedford Harbor area:

	SOURCE	AMOUNT REQUIRED
1.	Slightly Contaminated Sediments	3 gal
2.	Heavily Contaminated Sediments	10 gal
3.	Sediments Adjacent To A CSO Outfall	3 gal
4.	Sediments from a Local Salt Water Mudflat	3 gal

Representative samples of these sediments should be collected by others and express shipped prepaid to the Radian, Milwaukee laboratory. Radian personnel will provide instruction as to the recommended collection methods and sample shipment procedures. In addition, Radian will require a 55 gallon sample of New Bedford harbor salt water. This water should be collected by others and shipped by common carrier prepaid.

The slightly contaminated sediments may hold the best community since the microbes present would have had a chance over time to adjust to the presence of low level PCBs without being irrevocably shocked by a large dose.

Sediments from a high concentration area would be the most likely source for finding microbes which already were adjusted to the presence of PCBs before contacting the substance, and have had the time to potentially evolve further the ability to degrade the substance while being protected from competition from other nonresistant organisms.

A culture from near a CSO outfall in the area would be one which had been exposed to numerous organic compounds, a stressful environment and consecutive introduction of microbial communities. All of these factors would contribute to the evolution of a diverse microbial community which may contain the desired microorganisms.

The microorganisms from a mudflat or tidal pool are naturally exposed to a stressful environment as above, with the addition of having a drying and wetting cycle, which means that at least part of the time the system will be aerobic. Cultures from this area may be useful because of both the diverse populations and because PCB degradation has been associated with aerobic microorganisms. Mudpools and tidal basins have often been called the ocean's equivalent to a soil adsorption field for treatment of wastes from the land. A tidal pool in the PCB contaminated area would be ideal.

In addition, to these four salt water sources we will also collect seed from three other sources. These sources are from a fresh water environment, however many microorganisms can withstand either salinity environment. The three sources are:

1. Solids from a Functioning PCB Aerobic Sludge Digester

2. Samples of Activated Sludge Microorganisms

3. Commercial Microbial Cultures for the Degradation of PCBs

These cultures are readily available. Based on actual experience dissolved mineral content, or the salinity, in activated sludge water and in sludge digester water is frequently quite high. Thus it is reasonable to evaluate these sources of microorganisms also as potential seed.

3.0 WORKSCOPE

3.1 Task 1 - Project Plan Development

The objective of this task is the prepartion of a test plan for operation of the bench scale experiments. The test plan will include a complete discussion of the numbers and kinds of experiments to be performed, test parameters to be measured, sampling and analytical schedules, data reporting methods, the laboratory quality assurance/quality control plan and health/safety aspects of the project.

3.2 Task 2 - Culture Development

We propose to use biphenyl instead of PCBs as the initial selection mechanism because it is much safer to use. Several other authors have done this previously. All enzyme systems found to date capable of degrading PCBs can also degrade biphenyl. However the reverse is not always true, all enzyme systems that degrade biphenyl may not be capable of degrading all PCBs. For this reason, to verify the capability of the enhanced culture to degrade PCBs, PCBs of the mix of congeners found in the New Bedford Harbor will be used. Also, when sediment is introduced into cultures, reseeding from all the potential sources will be repeated.

The development of the enriched culture will be performed in two steps: (1) development of a culture able to degrade biphenyl in the presence of other substrates, and (2) using the first developed culture as seed, development of a PCB degrading culture using actual contaminated sediments as the carbon source.

All cultures will be developed in glass flasks which will either be shaken or bubble aerated. Five hundred milliliter flasks wrapped in aluminum foil to eliminate photodegradation will be used and filled with 150 ml of culture. Only salt water will be used in the cultures. The salt water will be prepared using sea salt or actual salt water from the New Bedford harbor, and will be supplemented with mineral nutrients if necessary. Aeration and mixing will be provided by shaking the flasks and continuously exchanging the air in the head space above the culture media.

3.2.1 Development of Biphenyl Cultures

The biphenyl cultures wil be developed in a media created by adding biphenyl, at 0.2 grams/liter and a salt water and raw sewage mixture to 6 flasks. The media will be seeded with each of the microbial seed sources initially and every other day for 10 days. A 5 day hydraulic detention time in the culture flasks will be controlled by daily letting solids in the flasks settle to the bottom and draining off 1/5 (30 ml) of clear supernatant. Then the same volume of the raw sewage and sea salt mixture will be added along with 0.1 grams of biphenyl.

General growth in the reactors will be monitored by determining the total and total volatile solids daily and oxygen uptake rates every five days. Once these have reached a steady state (after at least 15 days), the ability of the culture to degrade biphenyl will be determined.

Growth on biphenyl will be demonstrated by adding seed from the cultures to a second set of flasks containing nutrient enriched salt water and biphenyl. Growth on the biphenyl will be monitored by measuring the increase in the turbidity of the nutrient media of these verification cultures. To ensure that the turbidity is due to growth on the biphenyl and not other

substrates introduced with the seed, a second set of verification cultures will be developed using the first set as the seed source. If growth is not observed within five days in the test flasks, the seeding process of the set of sewage and biphenyl cultures will be repeated.

Once a successful culture has been developed, it will be maintained throughout the process as a backup in case of emergencies.

3.2.2 Development of PCB Cultures

The sediment PCB cultures will be developed following a similar process as the biphenyl cultures, however, sediment from the New Bedford Harbor will be used instead of the raw sewage. Again, biphenyl will be added daily but this time as a potential cometabolyte and a protected substrate source to boost the populations previously enriched. Seed for these cultures will come from the biphenyl/sewage cultures and from the different initial sources to insure that all sources of PCB degrading microorganisms have been added. Seeding will be performed every other day for the first 10 days and every 5 days thereafter.

Both the hydraulic and solids detention time will be controlled at 14 days. This will be done by drawing 1/14th of the culture while it is still mixing and filling with the same amount of fresh sediment and water. The solids level will be maintained at 1% solids. Again, general growth will be monitored by performing a mass balance on the solids and measuring oxygen uptake rates.

After 42 days (three solids detention times), the ability of the culture to degrade PCBs will be tested in two different ways. First, a ten day composite sample of the feed to the reactors and the effluent will be tested for PCB content. A difference would definitely be due only to biodegradation, as volatilization or sorption onto the reactor walls may have occurred.

Second, PCB biodegradation will be confirmed by adding seed from the sediment cultures to 6 flasks containing a fresh mixture of preaerated contaminated sediment, biphenyl, and nutrient enriched salt water. (Preaerated sediment is used to prevent death of the culture due to depletion of oxygen resulting from the initial oxygen demand of anaerobic sediments). These reactors will be operated in a batch mode for 14 days. In addition, control flasks will be set up containing the contaminated sediments and sea water, but also with sufficient sodium azide to inhibit any biological growth. (The proper dose of azide will be determined using total plate counts and different doses of azide in a preliminary test). A difference between the initial and final PCB levels in the test reactors above that found in the control reactors will be accepted as verification of PCB biodegradation.

If degradation is not found, the development process will be reviewed and repeated at least once before ending the experiment. Once a successful culture has been developed, it will be maintained throughout the project as a backup in case of emergencies.

3.3 Task 3 - Final Report

The final report will include complete documentation of the experimental program conducted and will consist of following elements:

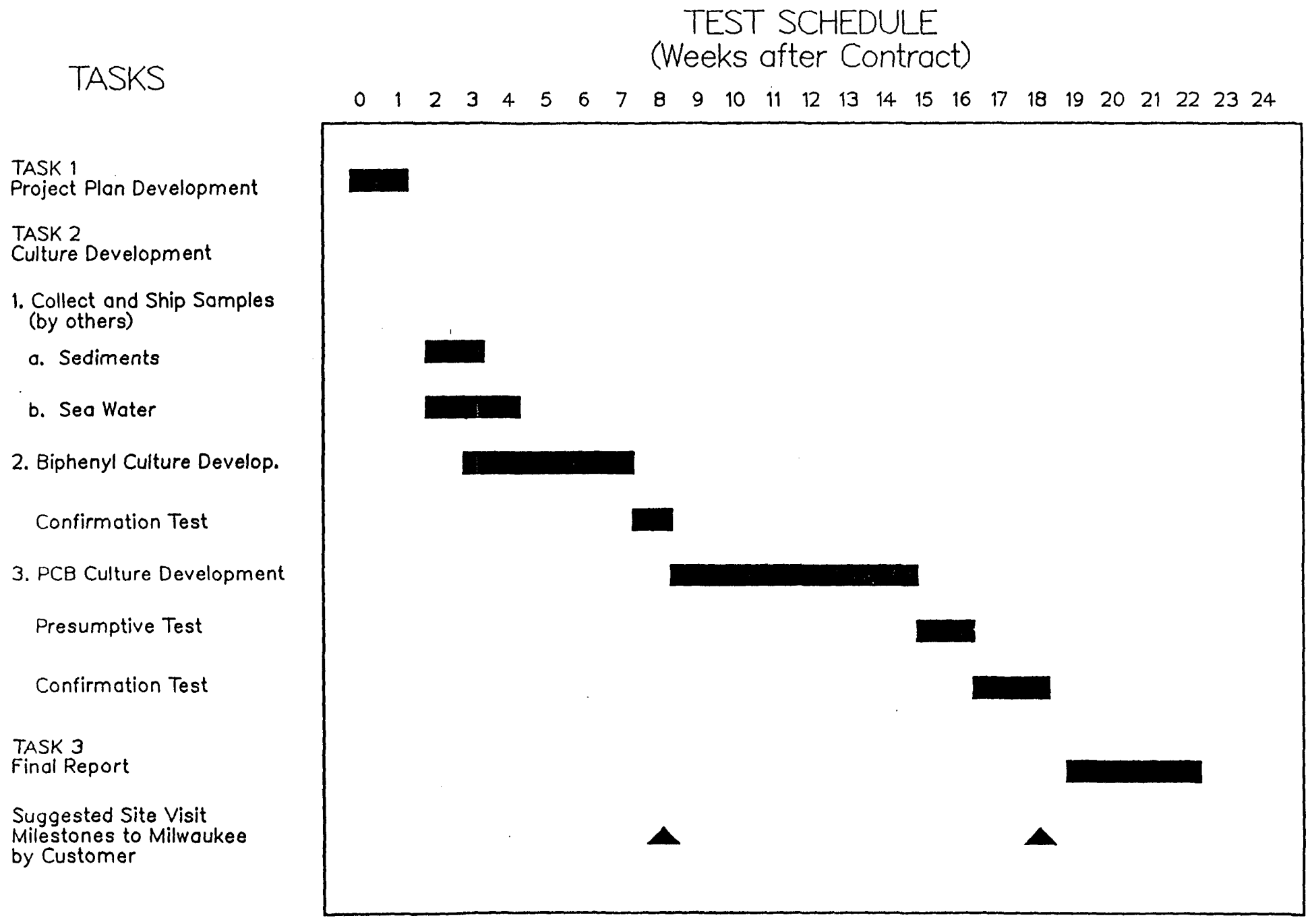
a. Results of data collection and analysis;

b. Basic design criteria and proposed operating conditions for future test phases;

c. Technical feasibility and economic evaluation of proposed treatment scheme;

d. Laboratory QA/QC results; and

e. Engineering conclusions and recommendations.


TEST SCHEDULE
(Weeks after Contract)
TASKS
0 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23 24
TASK 1
Project Plan Development
TASK 2
Culture Development
1. Collect and Ship Samples
(by others)
a. Sediments
b. Sea Water
2. Biphenyl Culture Develop.
Confirmation Test
3. PCB Culture Development
Presumptive Test
Confirmation Test
TASK 3
Final Report
Suggested Site Visit
Milestones to Milwaukee
by Customer

APPENDIX B

EXPERIMENTAL PROJECT PLAN

EXPERIMENTAL PROJECT PLAN

BENCH SCALE TESTING OF BIODEGRADATION TECHNOLOGIES FOR PCB'S IN NEW BEDFORD (MA) HARBOR SEDIMENT

SUBCONTRACT NO. 4236-LAB-0062

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October 5, 1987

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DEVELOPMENT OF AN ENRICHED SALTWATER MICROBIAL COMMUNITY CAPABLE OF BIODEGRADING PCBs SORBED TO HARBOR SEDIMENTS

EXPERIMENTAL PROJECT PLAN

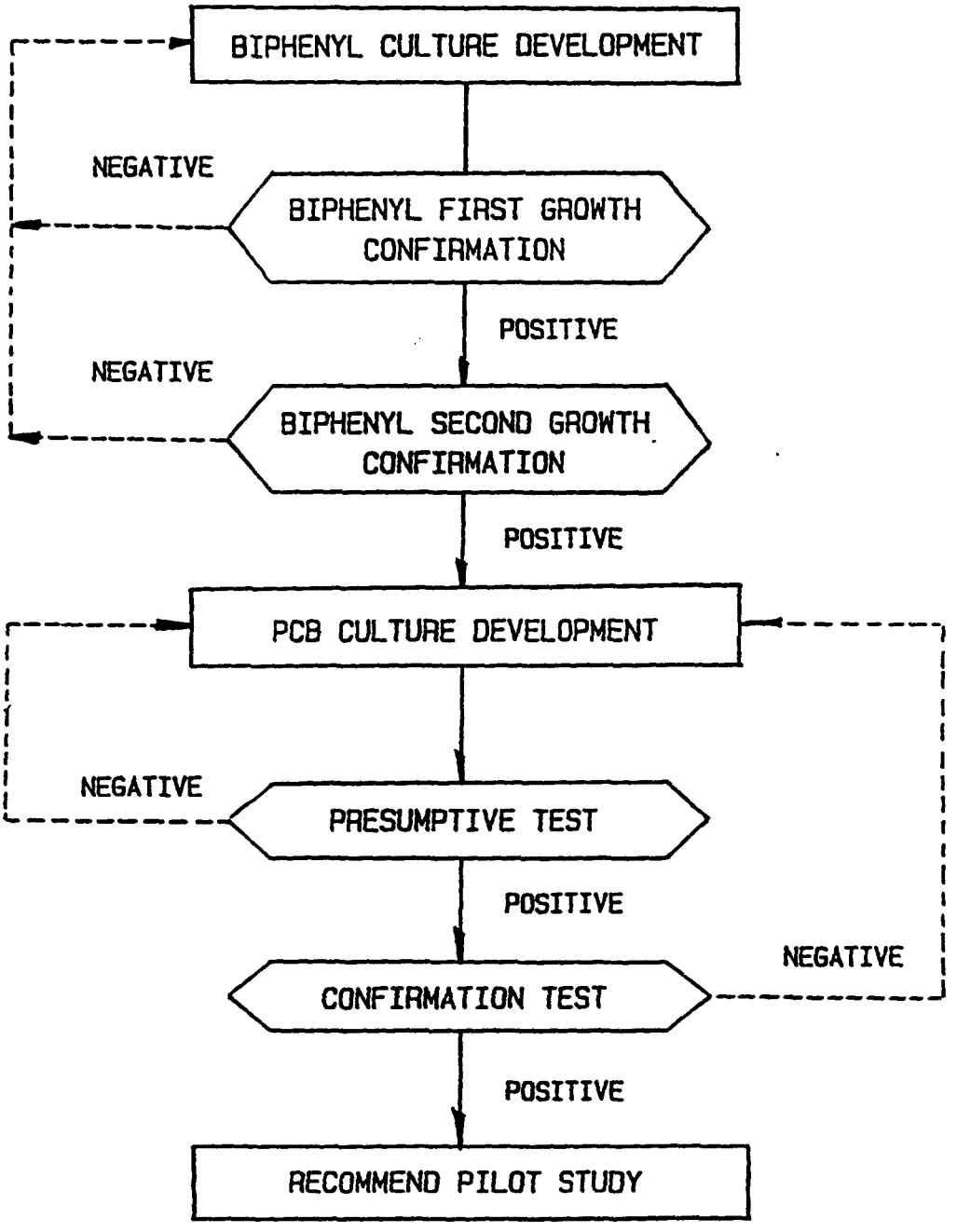
1.0 INTRODUCTION

The goal of this project is to develop a salt water microbial community capable of degrading PCBs and demonstrate this capability when the PCBs are sorbed to harbor sediments - specifically sediments from the harbor at New Bedford, Ma. We will use microbial communities taken from different areas in the harbor, microbial communities from sewage treatment works, from fresh water PCB degrading cultures, and from purchased cultures as seeds in the development of the mixed culture. Instead of PCBs we will use biphenyl to activate the selection mechanism for the microbial community for initial culture development. Then we will shift to actual PCB contaminated sediments for test purposes.

In logical sequence the experimental project plan first develops an enriched microbial culture from various inoculum sources using biphenyl and sewage as carbon substrates. This culture is then tested for its ability to grow on the biphenyl before proceeding to substitution of PCB contaminated sediment as the primary substrate. The ability of the enrichment culture to degrade PCBs will be tested first by a presumptive test based on the change in PCB concentration in continuously fed reactors. A confirmation test will follow which will include controls to determine if biological metabolism is solely responsible for PCB disappearance.

The overall project sequence and decision points are illustrated in Figure 1. Note that there are several milestones at which decisions are made whether or not to continue to the next step or to back up to a previous

FIGURE 1
MAIN PROJECT FLOW DIAGRAM



step. This approach is designed to maximize the opportunities to successfully develop a PCB degrading culture in the environment present in New Bedford harbor sediments.

A detailed discussion of the tasks required for this project follows. Each task is specified in five steps. The first describes the media used for culture development for the specific task. Initial conditions are then specified. Here the apparatus is described as well as quantities of materials initially present in the reaction flasks. The next subtopic, operations, describes on a daily basis the quantities of materials added to each reaction flask. Sampling and analysis requirements are then detailed. Finally, discussion of what action will be taken during the progress of each task is provided. The basis for making decisions and when action is to be taken is detailed.

Figures 2 and 3 present graphically the sequence of operation, monitoring and sampling activities which will be performed in the course of this project.

2.0 BIPHENYL CULTURES

The bench scale effort involving development of biphenyl cultures will be discussed in three sections: (1) development and acclimation of the microbial culture, (2) testing the ability of the developed culture to grow with biphenyl as the sole carbon source, and (3) testing of the culture grown on biphenyl to seed a second test culture.

2.1 Acclimation of the Microbial Culture

Before any testing of a mixed culture can be performed, acclimation of the culture to operating conditions must be undertaken. This acclimation period allows for a shifting in the population levels between species in the microbial community in the culture and the adjustment of the metablolism of the individual microorgansims to optimize competitive success.

The goal of this substep is to develop, or more accurately, allow a culture to develop which is capable of degrading biphenyl in the presence of alternative substrates.

The microorganisms in the culture not utilizing biphenyl will have to adjust to the presence of biphenyl. Those microbes capable of growing on biphenyl will have to produce the enzymes necessary, and adjust the quantity of production of the enzymes to correspond to the availability of biphenyl. This will allow them to optimize energetically the benefits of degrading the biphenyl and make themselves competitively successful in relation to the other microorganisms in the culture. To allow this to occur, the proper conditions must be established. These conditions are discussed below.

2.1.1 Media

The media used in this culture development will be primary sewage which has had sea salts added to bring its salinity to a level corresponding to ocean water. In addition, the following micronutrients will be added:

$MgSO_4 \cdot 7H_2O$ (0.05 g/L)
$FeSO_4 \cdot 7H_2O$ (0.01 g/L)
$CuSO_4 \cdot 5H_2O$ (0.01 g/L)
$CoCl_2 \cdot 6H_2O$ (0.01 g/L)

The sea salts are added because of the desire to operate in a saline environment. The trace elements are added as supplements. They are often used as cofactors or "hemmes" in microbial enzymes.

Biphenyl will be added to the reactors on a daily basis. It cannot be added as a constituent of the media as it will volatilize readily from the media, having a soluble half life in quiescent water in the range of 2-6 hours.

2.1.2 Initial Conditions

As previously identified, 6 reactors will be used in the substep. They will consist of 6 - 500 ml Erlynmeyer flasks, with standard stoppers which are wrapped with aluminum foil to prevent sorption to the stoppers. Air will be circulated in the head space of the reactor via glass tubes extending through the stoppers. Air flow rate will be approximately 100 ml/min. The reactors will be shaken at a rate sufficient to ensure aerobic conditions in the media.

The initial contents of the reactor will be 150 ml of media, 0.1 grams biphenyl, and seeds from each of the 7 microbe sources identified in Section 4.0. To seed each reactor 0.5 grams of sediment from each of the sediments from the New Bedford harbor and 1 ml of seed from the other three sources will be added.

2.1.3 Reactor Operations

After the initial setup of the reactors, they will be placed on wrist shakers which will be operated continuously. The reactors will be fed on a daily basis, 6 days a week, for the duration of the project step. The reactors will be operated in a batch draw and fill mode. That is, daily the wrist shaker will be turned off for approximately 15-30 minutes to allow solids in the reactors to settle. Then 30 ml of supernatant will be removed, replaced with 30 ml fresh media and 0.1 grams of biphenyl. On every sixth day of operation, double the amount of media and biphenyl will be exchanged to provide a food supply for the seventh day, on which no exchange will take place.

The 30 ml corresponds to a 5 day hydraulic detention time of the liquid phase. This is sufficient to supply a quantity of readily available organic food source in the sewage for the microorganisms and removal of metabolic byproducts which might inhibit continuous growth. A five day time was chosen so that the food supply would be sufficient to allow for survival of the microbe until they can shift to biphenyl as a food source, but low enough to not allow continuous growth on the organics in the sewage.

Given a 35 day maximum time period, 7.5 liters of culture and 21 grams of biphenyl will be required.

2.1.4 Sampling and Analysis

Growth on the biphenyl, sewage mixture will be monitored by biweekly sampling of the solids from each reactor, and by biweekly oxygen uptakes. Specifically, total solids and total volatile solids will be performed initially and on Tuesdays and Thursdays of each week on the effluent from the reactor. This corresponds to a maximum of 84 solids analysis over a 7 week period. Oxygen uptakes will be performed on Mondays and Wednesdays. Because 300 ml of media is required to perform on O_2 uptake test, the contents of 2 reactors will be combined for the analysis (resulting in 3 analysis per sampling day or 42 O_2 uptake measurements in 7 weeks). The sample will then be redivided into 150 ml fractions and returned to the culture reactors. Mixing pairs of flasks will allow cross seeding of cultures, increasing the opportunity for successful acclimation. Reactor pairs to be mixed will be selected randomly each sampling period.

In addition to these analyses, the total weight of each reactor will be measured prior to each feeding, and water will be added as needed to replace any evaporative losses.

The data from these analyses will be stored on Lotus spreadsheets, along with the sample day. The data will be plotted weekly to facilitate observation of the changes in analytical values.

2.1.5 Milestones and Decision Points

The key time in this project phase will be on days 5, 10, 15 and possibly 10, 25, and 30 of the reactor runs. At these times we will review the data for indication of microbial growth, such as an increase in the percent volatile solids or in O_2 uptake rates. If insufficient growth is observed, reseeding of each reactor will be undertaken. On day 15 the decision will be made whether or not to proceed to the biphenyl growth tests

or to reseed and continue acclimation. Day 15 was chosen since it corresponds to 3 detention times, which is the statistical time when 90% of steady state physical conditions are achieved. On day 35, if no growth has been observed, the biphenyl test will be undertaken irregardless and the decision to continue the project will be made based on the observed results.

2.2 Biphenyl Culture Testing

To determine if in the biphenyl acclimated cultures, a biphenyl degrading microorganism is present and competing successfully with the other miroorganisms, it is necessary to test the culture's ability to grow in a medium with biphenyl as the sole carbon source. The approach will be to add a 1 ml seed from the sewage/biphenyl culture to media composed of saltwater, nutrients, and biphenyl, and over a 5 day period, measure the turbidity. If an increase in turbidity is observed, this would indicate either microbial growth or microbial breakdown of the biphenyl. (It has been found that one of the breakdown products of biphenyl induces a color, usually yellow, to the media.) This seeding and growth will then be repeated, using the cultures as the new seed to ensure that the growth was on the biphenyl, and not on other carbon sources transferred in the seeding process.

2.2.1 Media

The media for use in this experimental substep will be made from organic carbon free water (deionized), sea salt, the micronutrients previously listed, and the following nitrogen and phosphorous sources:

K_2HPO_4 (1.3 gr/L)
KH_2PO_4 (0.82 gr/L)
$(NH_4)_2SO_4$ (1.0 gr/L)

The addition of the P and N will ensure that the growth is not limited by noncarbon sources. Also in these forms the nutrients will also provide a pH buffer in the range optimal for most microbial growth.

Again biphenyl will be added separately from the other media constituents.

2.2.2 Initial Conditions

The reactors used in this step will be duplicates of those used previously. Again 6 reactors will be operated, filled with 150 ml of media and 0.1 grams of biphenyl. Seeding of the first set of reactors will be performed by adding 1 ml of seed to each reactor. Seed for the second set of reactors will be taken from the cultures grown in the first set.

2.2.3 Operation

After seeding the reactors, the reactors will be shaken sufficiently to provide aerobic conditions. The reactors will be operated in a batch mode, no filling or drawing will take place. Biphenyl, 0.05 grams, will be added daily. Each set of 6 reactors will be operated for a minimum of 5 days, and a proposed maximum of 10 days.

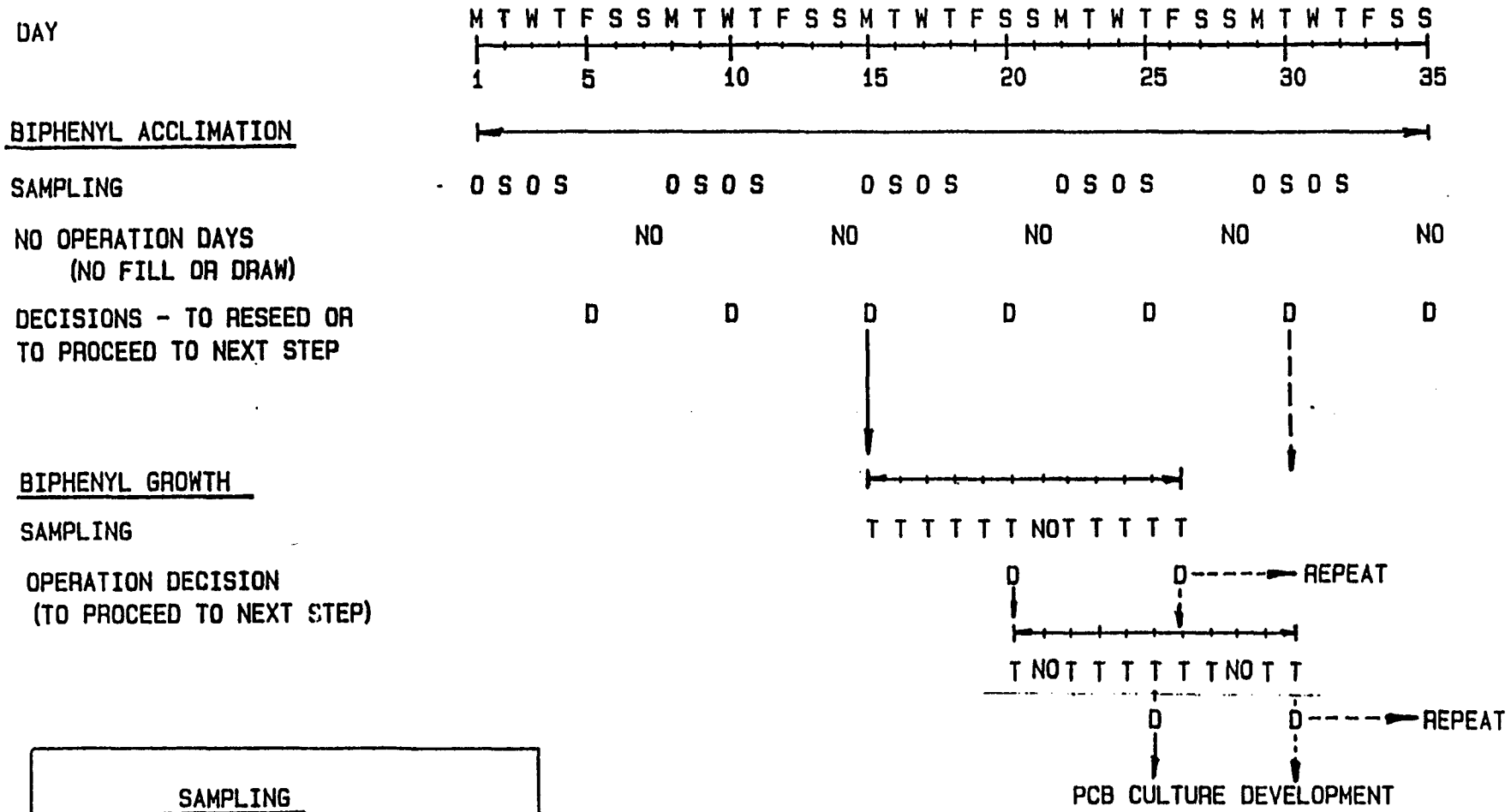
Given a 20 day maximum run time, 1.8 liters of media and 7 grams of biphenyl will be used.

2.2.4 Sampling and Analytical Schedule

The turbidity of each of the cultures will be measured 6 of every 7 days of reactor run (Monday thru Saturday). The sample used to determine turbidity will not be harmed and will be returned to the reactors. The weight of the reactor will also be measured on the same schedule to check for evaporative losses.

The turbidity data, and the sampling time will be stored in a Lotus database and plotted to facilitate observation of changes in turbidity. A maximum of approximately 108 turbidity analyses will be performed.

FIGURE 2
DEVELOPMENT OF BIPHENYL CULTURES



SAMPLING

O = OXYGEN UPTAKE
S = TOTAL & TOTAL VOLATILE SOLIDS
T = TURBIDITY
NO = NO OPERATION

2.2.5 Milestones and Decision Points

The critical times in the testing of growth on biphenyl will be day 5 and day 10 of each test set. On day 5, set #1, the turbidity data will be reviewed. If no increase in turbidity is seen, reseeding will take place on this day. If an indication of growth is observed, but is not statistically significant (based on an analysis of variance), the decision will be made to either reseed or continue for 5 days (or both, using 3 reactors for each condition). If no change in turbidity is measured, the reactors will be reseeded.

On day 10, set #2, (or if turbidity had increased by day 5) the next decision will be made. If turbidity has increased, the second set of culture tests will be initiated. The decision steps for the second set of tests will follow that presented for the first set regarding timing and reseeding.

If growth is indicated in these reactors after a maximum of ten days, the initiation of PCB cultures will be undertaken. If growth is not observed, the decision will be made to continue the acclimation of the sewage/biphenyl reactors for a second 15 days and retest the ability to grow on biphenyl. (Note that these reactors were still under operation during this testing period.)

If after the second testing period, no growth is indicated, we will present our findings to EBASCO and discuss with them possible future actions.

3.0 DEVELOPMENT OF PCB CULTURES

Once an acclimated culture is developed, the initiation of the development of a PCB degrading culture can be started. The sewage/biphenyl culture will be used as a seed for this step. The culture will be developed using the actual sediments from the New Bedford harbor with the nutrients

supplemented using micronutrients and biphenyl. This culutre development is divided into 3 substeps:

A. Culture Acclimation
B. Presumptive Testing
C. Confirmed Degradation

The goal of this project step is to acclimate the developed culture and demonstrate the ability of this culture to degrade PCBs.

3.1 Culture Acclimation

It is necessary to reacclimate the sewage/biphenyl culture to the sediment/biphenyl mix for two main reasons, a change in carbon sources, and a change in the operating prarmeters of the reactors.

The change in carbon sources is a result of shifting from sewage to sediments. In sewage, most of the carbon sources are soluble organics and are readily degradable. In sediments, the carbon sources are mainly relatively insoluble and a major fraction are sorbed to solids. The microbial community capable of degrading sewage substrates may not contain the same population levels or distribution of microbes as the sediment substrate.

The change in operating conditions to a 14 day solids and hydraulic retention time will also effect the microbial community because of two main reasons. First, the food/microorganism ratio will be diffeent at least initially. Second, the microorganisms will have a longer period to degrade the substrates. The increase was made because prior studies have shown that microbes degrade PCBs much slower than biphenyl. It was felt after 14 days sufficient PCB degradation would have occurred to be statistically significant.

3.1.1 Media

The media used in this project phase will be from two sources of sediments collected from a PCB hot spot (greater than 3000 gpm), and from a

less contaminated spot (less than 1000 ppm). (This is done to provide more information on the limits of the biodegradation process.) Both sediments will be diluted to 1% solids using sea water collected from the New Bedford harbor. To this the micronutrients will be added. If upon testing of this mix, nitrogen and phosphorous levels are found to be potentially limiting, these nutrients will also be introduced.

Biphenyl will again be added independently of the media, due to its volatility.

3.1.2 Initial Conditions

Six reactors will be used in this substep. One liter Erlynmeyer flasks will be used instead of 500 ml flasks, otherwise the same design will be used as in the biphenyl culture development. Each reactor will be filled with 300 ml of media and 0.2 grams of biphenyl. (The increase in size and media volume will provide for larger samples for analysis.) Four reactors will be fed with media made up from the hot spot sediment, two from the less contaminated sediment.

The reactors will be seeded using the sewage/biphenyl cultures. Ten milliliters of culture will be added to each reactor. In addition, the other sources of liquid cultures, commercial culture, activated sludge and culture from a PCB degrading aerobic reactor will be added. The seeding will be performed at the beginning of the second day of operation so that the media will have had sufficient time to become aerobic.

Initial seeding will be performed every other day for the first 10 days of reactor operation.

3.1.3 Operation

These reactors will be operated in a draw and fill mode (6 days out of every 7), similar to that used for the biphenyl culture acclimation except that the reactor solids will not be settled prior to draw. Given a 300 ml volume and a 14 day detention time, 22 ml will be drawn on the first to fifth

day of each 7 day operation schedule. On the sixth day 44 ml will be drawn. The volume will be replaced with the 1% solids media and 0.1 grams of biphenyl. The reactors will be operated for 42 days (three detention times) in this mode. For the 4 high PCB reactors, 5.0 liters of media will be needed. For the 2 lower PCB reactors, 2.5 liters of media will be needed. Thirty grams of biphenyl will be needed.

3.1.4 Sampling and Analysis Schedule

In that during this period, the goal is only to acclimate the microbial culture, no testing of the PCB degradation is necessary. General opeation variables will be monitored, including solid levels, O_2 uptakes, and O_2 levels. Both total and total volatile solids will be analyzed on every Tuesday and Thursday, using the effluent from the draw and fill operation. For the 42 day operation, 72 total solids and total volitile solids will be performed.

Dissolved oxygen levels will be measured on Mondays and Wednesdays, oxygen uptake rates will be performed on Wednesdays. Seventy-two dissolved oxygen levels and 36 uptake rates will be determined. The resulting data will be collected and analyzed for trends using Lotus spreadsheet and graphics.

3.1.5 Milestones and Decision Points

The decision points for this project phase will be every 14 days. At this time, the data will be reviewed and the need for repeated seeding will be determined. On day 42 the decision will be made whether to proceed to the presumptive testing.

3.2 Presumptive Testing

This project substep involves the measurement of the change in the mass of PCBs in the sediment reactors over a 10 day period. The test is considered presumptive in that it measures only the change in PCB mass, and does not demonstrate that the disappearance is due to biodegradation.

3.2.1 Media

3.2.2 Initial Conditions

3.2.3 Operation

This project substep will simply involve continuing the operation of the acclimation reactors for a 10 day period beyond the initial 42 days. This will require 1.8 liters of the high PCB sediment media, 0.90 liters of the lower PCB sediment media, and 6 grams of biphenyl. (One half of the feed will be for composite analysis.)

3.2.4 Sampling and Analysis

During this project step samples for PCB analysis of the feed to the reactors and effluent from the reactors will be taken for shipment to Jordan Laboratories and for in-house analysis.

To determine the PCB levels and distribution of PCB congeners in the feed to the reactors a 10 day composite sample of the feed to each reactor will be taken. Specifically, 22 ml of media will be added to the reactors and to a composite sample for each reactor. At the end of the 10 day period the volume of composite feed will be 220 ml. Of this 40 ml will be sent to Jordan for analysis (sufficient for 2 homologue tests and 2 680 tests). The remaining volume will be used for the in-house analysis for PCBs and for total and total volatile solids. This corresponds to 14 PCB analyses by Jordan, 2 PCB analysis in-house (one on a high PCB media and one on a lower PCB media), and 6 total and total volatile solids.

To determine the corresponding information in the reactor effluents a second set of composites will be taken. The draw from each of the reactors will be composited separately for the 10 day period (220 ml in each of 6 composites). Again 40 ml will be sent to Jordan for analysis from each composite, and the rest used for in-house analysis for PCBs and solids. This

again corresponds to 24 PCB analysis by Jordan, 2 PCB analysis in-house, and 6 total and total volatile solids. The reactor will be weighed daily to determine evaporative losses.

The data from these analyses will be stored in Lotus spreadsheets. For each PCB sample and analysis the total PCB levels and the distribution of homologues will be stored. Data analysis will involve homologue by homologue comparison and change in total PCB levels.

3.2.5 Milestones and Decision Points

The decision point in this step will be upon an initial analysis of the PCB data and development of mass balances of PCBs in the reactors. If a reduction of PCBs is found and the reduction is greater in the lower chlorinated biphenyls than overall, the analysis of confirmation test sample will be initiated. (To minimize project time, we will perform the presumptive test immediately.) If no significant or constant change is seen, a second set of composite samples could be taken.

3.3 Confirmation of PCB Biodegradation

To determine if PCBs are actually biodegraded and not sorbed to reactor walls or volatilized, batch reactors which will be seeded using the cultures in the confirmation reactors will be operated for 14 days. Along with these reactors, two other reactors will be operated in which biological growth will be eliminated. Biological decomposition will be considered demonstrated if the reduction in PCB levels is greater in the biological reactors than in the control reactors. In addition, further evidence for biological decomposition will be in any differences in the distribution of the homologues between the biological reactor samples and the control reactor samples.

3.3.1 Media

The media used in the confirmation reactor test will be the same as for the presumptive tests and the acclimation reactors.

3.3.2 Initial Conditions

Eight one liter reactors will be used for this project step. Six reactors will be used for biological degradation and 2 as controls. Of the 6 reactors, 4 will contain 300 ml of high PCB sediment media and 2 will contain 300 ml of lower PCB sediment media. One control reactor will contain 300 ml of high PCB sediment sludge, the other 300 ml of the lower PCB sediment sludge. To all 8 reactors, 0.2 grams of biphenyl will be added. The control reactors will also contain a biological toxin to prevent biological growth.

Seeding of the reactors will take place after the reactors have been aerated for 24 hours. This is to insure that the reactor media is aerobic prior to introduction of seed. Ten ml of seed culture will be added to each of the 6 biological reactors. The seed will be taken from the presumptive test reactors. The seeding will be repeated at the beginning of day 5 to insure successful seeding. Two liters of high PCB sediment media, 1 liter of lower PCB sediment media, and 2 grams of biphenyl will be needed.

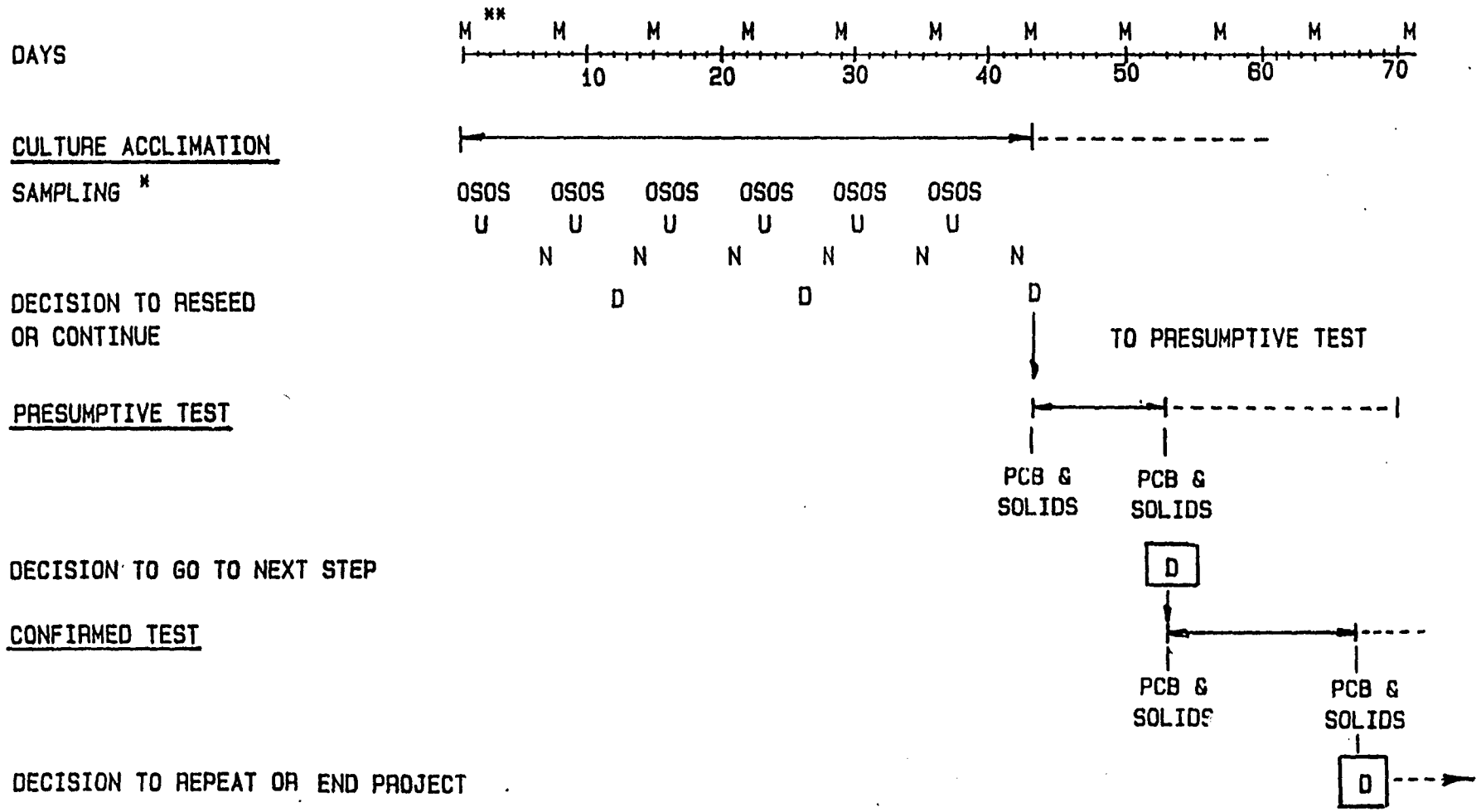
3.3.3 Operation

The operation of the confirmation reactor will consist of aerating and stirring the reactors for a 14 day period. No drawing or filling will take place. However, 0.1 grams of biphenyl will be added daily to the reactors.

3.3.4 Sampling and Analysis

These reactors will be sampled for PCBs and solids levels at the beginning and end of the 14 day period. For sampling of the initial conditions, 8-100 ml samples of media split from the media mixed for each reactor will be taken. Of this, 8-40 ml samples will be sent to Jordan for PCB analysis, the rest used for in-house PCB and solids analysis. Two in-house PCB analyses, one on a high PCB sediment media, and one on the lower PCB sediment media will be performed. Total and total volatile samples will be determined on each of the 8 reactor samples. Jordan will perform 24 PCB

FIGURE 3
DEVELOPMENT OF PCB CULTURE



* SAMPLING KEY: O=DISSOLVED OXYGEN; S=TOTAL AND VOLATILE SOLIDS; U=OXYGEN UPTAKE ; N=NO OPERATION

** MONDAY

analyses, Radian 2 PCB analysis, and 8 sets of solids analyses on the inital reactor media.

For sampling of the reactor contents at the end of the 14 day run, the entire contents of the reactors will be available for analysis. Again, 8 samples of 40 ml each will be sent to Jordan for PCB analysis. Two in-house PCB analyses will be performed, and 8 sets of solids analyses.

The data handling will be equivalent to that performed on the presumptive test. When analyzed, the amount of change found in the PCB levels in control reactors by homologue will be subtracted from the amount of change in the biological PCB levels in the reactors. The remaining "change" in PCB homologue will then be examined for overall reduction and individual homologue reduction.

3.3.5 Milestones and Decision Points

The analysis of the change in PCBs in this confirmation test will determine the success of this project.

4.0 MICROBIAL AND PCB SUBSTRATE SOURCES

We will develop the culture from seven sources of microorganisms, each of which may contain microorganisms capable of degrading PCBs. We will use the idea, as has been found in developing communities for treating other recalcitrant compounds, that nature has already performed a great deal of selection and sorting. That is, if one wants to find a PCB degrading microorganism capable of doing so in a certain environment, it is best to look in that environment or a similar one where the PCBs are and have been present for a length of time.

We propose to use four sources of microbes from the New Bedford harbor area:

SOURCE	AMOUNT REQUIRED
1. Slightly Contaminated Sediments	1 gal.
2. Heavily Contaminated Sediments	2 gal.
3. Sediments Adjacent to a CSO Outfall	1 gal.
4. Sediments from a Local Salt Water Mudflat	1 gal.

In addition to providing a microbial inoculum, sources 1 and 2 will provide the PCB substrate to be used in section 3.0.

Representative samples of these sediments should be collected by others and express shipped prepaid to the Radian, Milwaukee laboratory. Radian personnel will provide instruction as to the recommended collection methods and sample shipment procedures. In addition, Radian will require a 55 gallon sample of New Bedford harbor salt water. This water should be collected by others and shipped by common carrier prepaid.

The slightly contaminated sediments may hold the best community since the microbes present would have had a chance over time to adjust to the presence of low level PCBs without being irrevocably shocked by a large dose.

Sediments from a high concentration area would be the most likely source for finding microbes which already were adjusted to the presence of PCBs before contacting the substance, and have had the time to potentially evolve further the ability to degrade the substance while being protected from competition from other nonresistant organisms.

A culture from near a CSO outfall in the area would be one which had been exposed to numerous organic compounds, a stressful environment and consecutive introduction of microbial communities. All of these factors would contribute to the evolution of a diverse microbial community which may contain the desired microorganisms.

The microorganisms from a mudflat or tidal pool are naturally exposed to a stressful environment as above, with the addition of having a drying and wetting cycle, which means that at least part of the time the system will be

aerobic. Cultures from this area may be useful because of both the diverse populations and because PCB degradation has been associated with aerobic microorganisms. Mudpools and tidal basins have often been called the ocean's equivalent to a soil adsorption field for treatment of wastes from the land. A tidal pool in the PCB contaminated area would be ideal.

In addition, to these four salt water sources we will also collect seed from three other sources. These sources are from a fresh water environment, however, many microorganisms can withstand either salinity environment. The three sources are:

1. Solids from a Functioning PCB Aerobic Sludge Digester
2. Samples of Activated Sludge Microorganisms
3. Commercial Microbial Cultures for the Degradation of PCBs

These cultures are readily available. Based on actual experience dissolved mineral content, or the salinity, in activated sludge water and in sludge digester water is frequently quite high. Thus it is reasonable to evaluate these sources of microorganisms also as potential seed.

5.0 LOCATION AND DESCRIPTION OF FACILITIES

All bench scale tests will be performed at the Radian Environmental Management facility located in Milwaukee, Wisconsin. All sediment and sea water samples should be sent to:

Radian Corporation
5103 W. Beloit Road
Milwaukee, Wisconsin 53214

Attention: C. S. Applegate

PCB analysis using GC/MS procedures (EPA Method 680) to identify homologues will be performed at Radian Corporations's Analytical Services Laboratory located in Austin, Texas. All other analyses described in this project plan will be performed in the Milwaukee laboratories.

Radian Milwaukee is based in a three building complex. This facility provides office and laboratory space for about 85 engineers, scientists and staff. Major technical facilities and equipment are described below.

LABORATORIES

Radian Environmental Management Laboratories

Most of the analytical and bench test work performed for this project will be performed in this laboratory. This laboratory consists of several work areas:

Analytical Laboratory - This 1,020 sq. ft. laboratory is used to receive and log in samples, split the samples for preservation, perform sample preparation steps such as digestion, distillation or concentration and perform most of the wet chemistry work. The laboratory has 152 feet of bench space including 19 feet of fume hood area used for digestions, distillations, Kuderna-Danish concentration and performance of other analyses involving hazardous materials. The room is supplied with hot, cold and dionized water, gas, compressed air, vacuum and 110 V and 220 V circuits. A Milli-Q Water Purification System is used to convert deionized water to reagent grade water for use in analytical procedures. The room contains a Heinecke glassware washer, an autoclave, refrigerator, BOD incubators, ovens and muffle furnace. A special vent is installed in the ceiling above the ovens and furnace to remove fumes and heat.

Instrument Room - Instrumental analyses are performed in this 660 ft^2 room containing 48 feet of desk height bench space and 48 feet of normal height (36") bench space. The laboratory is supplied with hot, cold and deionized water, gas, vacuum, compressed air and 110 V and 220 V circuits. A small fume hood vent is used to remove fumes from the atomic absorption spectrophotometer.

Balance Room - A 132 ft^2 room connects the Instrument Room and Analytical Laboratory. It contains three marble balance tables placed on a section of floor isolated from the remainder of the building with vibration absorbing material. The room is supplied with 110 V circuits.

Process Test Laboratory - Most of the bench scale process test work is performed in this 250 sq. ft. laboratory equipped with 34 feet of bench space. The laboratory is equipped to test treatment processes, perform laboratory scale experiments and perform a limited amount of analyses. The room is equipped with a refrigerator for temperature sensitive reagents and equipment needed to perform and support the treatment process testing activity. The laboratory is supplied with hot and cold tapwater, deionized water, gas, vacuum, compressed air and 110 V circuits.

Bench Test Room - This 432 sq. ft. room is used to perform small scale pilot tests and bench tests too large for the Process Test Laboratory. It has a 12 foot ceiling, is supplied with hot, cold and deionized water, compressed air, gas, vacuum, 110 V, 220 V and 440 V circuits and has a 54 foot long trench drain along a portion of the walls to receive wastewater from bench and pilot tests. Brackets along one wall are used to support tall test equipment such as lysimeters. The room contains a freezer, Wiley mill, a large dryer for vegetation samples and a Heinecke washer for cleaning sample bottles. Connected to this room is an 8 x 10 foot walk-in refrigerator capable of holding several 55 gallon drums of wastewater in addition to serving as the primary storage area for smaller samples. The refrigerator has a table, compressed air and 110 V outlet for performing bench tests in a cold environment. Adjacent to the bench test room is an equipment storage room containing a work bench and an 8 x 10 foot freight elevator connected to a truck unloading dock.

Chemical and Materials Research Laboratory - Research in the formulation of new chemical products and the application of plastics to new areas is performed here. Work includes the analysis of organic chemicals using FTIR and HPLC instruments.

Materials Analysis Laboratory - Analysis of ferrous, nonferrous and plastic materials used in production and research work is performed using classical wet chemistry methods, x-ray emission spectrometry and the electron microscope.

Endurance testing Laboratory - Static, dynamic and endurance testing is performed on Radian products and materials in development using over 70 standard and specifically built testing machines.

EQUIPMENT

A partial list of treatment processes that can be bench tested in the Radian Environmental Management laboratory is presented in Table 1. A list of bench scale equipment for performing treatability tests is presented in Table 2. Major items of analytical equipment used by the Radian laboroatories are listed in Table 3.

ENGINEERING AND DESIGN FACILITIES

The Radian Milwaukee Group contains complete modern engineering and design facilities including a computer center and CAD/CAM eqipment.

TECHNICAL LIBRARY

A technical library with direct access to computerized databases facilitates the performance of literature searches.

TABLE 1

AVAILABLE BENCH TEST PROCESSES

Solids Separation

(1) Gravity flotation
(2) Gravity sedimentation
(3) Dissolved-air flotation
(4) Filtration

Chemical Treatment

(1) Screen chemicals for feasibility and order of addition
(2) Chemical dosage
(3) Quick mix time
(4) Flocculation time
(5) Sedimentation or flotation tests

Sludge Thickening or Dewatering

(1) Gravity thickening
(2) Flotation thickening
(3) Vaccum filtration
(4) Centrifugation
(5) Belt filtration
(6) Filter press, plate & frame and diaphragm

Biological Treatment

(1) Activated sludge
(2) Nitrification
(3) Denitrification
(4) Anaerobic digestion

Advanced Waste Treatment

(1) Tertiary treatment
(2) Filtration
(3) Activated carbon adsorption
(4) Reverse osmosis
(5) Ozonation
(6) Electrodialysis
(7) Alkaline chlorination
(8) Ion exchange

Contaminated Soil Treatment

(1) Leaching
(2) In-situ chemical treatment
(3) Soil scrubbing

Solid Waste

(1) Lysimeter tests

TABLE 2

PARTIAL LIST OF TREATMENT PROCESSES

Water and Wastewater Treatment Equipment

Flocculation apparatus, Phipps & Bird
Sedimentation test equipment
Dissolved-air flotation test equipment
Multi-media filter
Micro-screen test apparatus
Carbon isotherm test equipment
Carbon column test equipment
Reverse osmosis test apparatus
Pluggage index test equipment
Activated sludge reactors
Arthur respirometer
Oxygen uptake apparatus (air stripping)
Ozonator, Orec Model 03V5-0

Chemical Dosage Testing

Zeta-meter
Capillary Suction Time (CST) apparatus

Sludge Treatment and Testing Equipment

Flotation thickening equipment
Gravity thickening equipment
Filter leaf and Buchner funnel test equipment
Centrifuge test equipment
Inclined scraper test equipment
Anaerobic digestion apparatus
Roll skimmer test apparatus

Soil, Sediments and Solid Waste Testing Equipment

Lysimeters
Laboratory contaminated soil scrubber (extractor)
Laboratory in-situ contaminated soil treatment test equipment
EP Toxicity test extractor
Sediment Oxygen Demand (SOD) chamber

Recorders, Controllers and Meters Used in Treatment Testing

Gas flow meters
Water meters
pH
ORP
Temperature
Dissolved oxygen
Pressure

TABLE 3

MAJOR ITEMS OF ANALYTICAL EQUIPMENT

Atomic Absorption Spectrophotometer
Perkin Elmer Model 403

- Background correction
- Electrodeless discharge lamp power source
- Flameless mercury analyzer
- Model HGA2200 graphite furnace
- Delves cup attachment

Autoanalyzer, Technicon AAII Single Channel

- Phosphorus module
- Ammonia module
- Nitrate/nitrite module

Bacteriological Equipment

- Barnstead automatic autoclave
- Air and water incubators
- Biological hood & glovebox with sub-virus air filtration capability

Balances

- Mettler Model H-5 analytical
- Mettler Model H-10 analytical
- Mettler Model H-20 semimicro

BOD Incubators, four, approx. 13 cu. ft. each

Centrifuges

- Sorvall SS-3 Superspeed
- Sorvall GLC-2
- IEC Model V, Size 2

Conductivity Meters, Two

Data Reduction

- Monroe Model 1766 desktop programmable calculator
- Perkin Elmer Sigma - data system
- Time-Sharing computer facilities

Dissolved Oxygen Probes

- YSI, two, BOD type
- Orion, BOD type
- Rexnord Instrument Products, two, field type

Electron Microscope

- Micro-probe analysis capability

Flash Point Tester, closed cup

EP Toxicity Test Extractor

Fluorometer, Turner Model II

Burrell Wrist Action Shaker - (2)

Gas Chromatographs

- Barber-Colman Series 5000, with:
  - thermal conductivity detector
  - dual flame ionization detectors, and
  - electron capture detector (Ni^{63})
- Perkin Elmer Model 3920 B, with:
  - thermionic (nitrogen/phosphorus) detector,
  - dual flame ionization detectors, and
  - electron capture detector (Ni^{63})
- Tracor Model 540, with:
  - Hall detector
  - photoionization detector
  - Purge-and-Trap

Ion Specific/pH meters, six, with specific ion electrodes

Liquid Chromatograph, Waters with:

- Absorbance detector
- Differential refractometer detector

Melting Point Apparatus

Microscopes

- Bausch & Lomb
- Bausch & Lomb binocular, movable stage, phase contract, camera
- American Optical stereoscopic microscope

Millipore Mill-Q Water Purification System for Research Grade Water

Ovens, five drying (103°C and two muffle furnaces (550°C)

Parr Adiabatic Calorimeter

Sample Storage

- Freezer, 13.5 cu. ft.
- Refrigerators, three, approx. 13 cu. ft. each
- Walk-in cooler, 8 ft. x 10 ft.

Spectrophotometers

- Bausch & Lomb Model 88
- Bausch & Lomb Model 100
- Nicolet FTIR

Surface Tension Apparatus, Cenco-duNuoy

Total Organic Carbon Analyzer, Dohrmann - Xertex

Turbidimeters, three Hach Model 2100 units

Ultrasonic Sieve Shaker

Viscometers, Brookfield, Stormer and Ostwald

Washer-Heinecke Typhoon Laboratory Washer

Wiley Mill

X-ray Emission Spectrometer

APPENDIX C

QUALITY ASSURANCE PROJECT PLAN

QUALITY ASSURANCE PROJECT PLAN

DEVELOPMENT OF AN ENRICHED SALTWATER MICROBIAL COMMUNITY CAPABLE OF BIODEGRADING PCB's SORBED TO HARBOR SEDIMENTS

EBASCO Contract No. 4236-LAB-0062

Radian No. 291-012-29-39

Prepared for: EBASCO Services, Inc.
2000 15th Street N.
Arlington, VA 22101
Mr. N. A. Fiscinia

Prepared by: Radian Corporation
5103 W. Beloit Rd.
Milwaukee, Wisconsin 53214

April 14, 1988

INTRODUCTION

This Quality Assurance Project Plan (QAPP) covers the sampling and analysis of samples obtained during bench scale treatability of PCB contaminated saltwater sediments. All bench scale tests will be performed at Radian Environmental Management facility located in Milwaukee, Wisconsin.

The goal of this project is to develop a saltwater microbial community capable of degrading PCB's and demonstrate this capability when the PCB's are sorbed to harbor sediments - specifically sediments from the harbor at New Bedford, MA. Microbial communities taken from different areas in the harbor, microbial communities from sewage treatment works, and from fresh water PCB degrading cultures will be used as seeds in the development of the mixed culture. Instead of PCB's, biphenyl will be used to activate the selection mechanism for the microbial community for initial culture development. The final phase will shift to actual PCB contaminated sediments for test purposes.

The data obtained are to be used for evaluation of remedial alternatives in the New Bedford Harbor Feasibility Study. The data quality objectives (DQOs) are premised on obtaining data that are adequate for screening remedial alternatives which utilize biodegradation as a treatment technology and DQOs are to be consistent with Analytical Level III as described in Data Quality Objectives for Remedial Response Activities, Volume I - Development Process, US EPA Publication 540/6-87/003A; March 1987.

All bench scale tests will be performed at the Radian Environmental Management facility in Milwaukee, Wisconsin. PCB analysis using GC/MS procedures (EPA Method 680) to identify homologues will be performed at Radian Corporation's Analytical Services Laboratory located in Austin, Texas. All other analyses will be performed at the Milwaukee laboratories. Revise last sentence to read . . .Additional PCB analyses will be managed through the U.S. EPA Contract Laboratory Program (CLP), in accordance with CLP protocols. These samples will be analyzed using EPA Method 680 and EPA Method 608.

This Quality Assurance Project Plan is based on the guidelines and specifications as described in "Interim Guidelines and Specifications for Preparing Quality Assurance Project Plan", QAMS 005/80, December 29, 1980.

TITLE PAGE

DETAILED QUALITY ASSURANCE PLAN

DEVELOPMENT OF AN ENRICHED SALTWATER MICROBIAL COMMUNITY CAPABLE OF BIODEGRADING PCB's SORBED TO HARBOR SEDIMENTS

EBASCO CONTRACT NO. 4236-LAB-0062

Submitted to: EBASCO Services, Inc.
2000 15th Street N.
Arlington, VA 22101
Mr. N. A. Fiscinia

Submitted by: Radian Corporation
5103 W. Beloit Road
Milwaukee, Wisconsin 53214

April 14, 1988

Radian Program Director
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Radian Project Manager
C. S. Applegate

Radian QA Officer
C. J. Crosby

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TABLE OF CONTENTS

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B. Chantry - University of Wisconsin

C. Applegate - Radian, Milwaukee - Project Manager

G. Huibregtse - Radian, Milwaukee - Project Engineer

C. Crosby - Radian, Milwaukee - QA Officer

PROJECT DESCRIPTION

The goal of this project is to develop a saltwater microbial community capable of degrading PCB's and demonstrate this capability when the PCB's are sorbed to harbor sediments - specifically sediments from the harbor at New Bedford, MA. We will use microbial communities taken from different areas in the harbor, microbial communities from sewage treatment works, from fresh water PCB degrading cultures, and from purchased cultures as seeds in the development of the mixed culture. Instead of PCB's we will use biphenyl to activate the selection mechanism for the microbial community for initial culture development. Then we will shift to actual PCB contaminated sediments for test purposes.

In logical sequence the experimental project plan first develops an enriched microbial culture form various inoculum sources using biphenyl and sewage as carbon substrates. This culture is then tested for its ability to grow on the biphenyl before proceeding to substitution of PCB contaminated sediment as the primary substrate. The ability of the enrichment culture to degrade PCB's will be tested first by a presumptive test based on the change in PCB concentration in continuously fed reactors. A confirmation test will follow which will include controls to determine if biological metabolism is solely responsible for PCB disappearance.

The overall project sequence and decision points are illustrated in Figure 1. Note that there are several milestones at which decisions are made whether or not to continue to the next step or to back up to a previous step. This approach is designed to maximize the opportunities to successfully develop a PCB degrading culture in the environment present in New Bedford harbor sediments.

The project is divided into two phases. The first phase, biphenyl culture development, will consist of the following subphases:

1. Culture Acclimation
2. Biphenyl Culture Testing

This phase will be used to develop the microbial community needed to seed the actual PCB contaminated sediment. Within this phase, total solids, total volatile solids, salinity, and oxygen uptake rate will be determined. See Figure 2 for sampling schedule.

The second phase has three subphases associated with it. They are:

1. Acclimation
2. Presumptive Testing
3. Confirmed Degradation of PCB's

FIGURE 1
MAIN PROJECT FLOW DIAGRAM

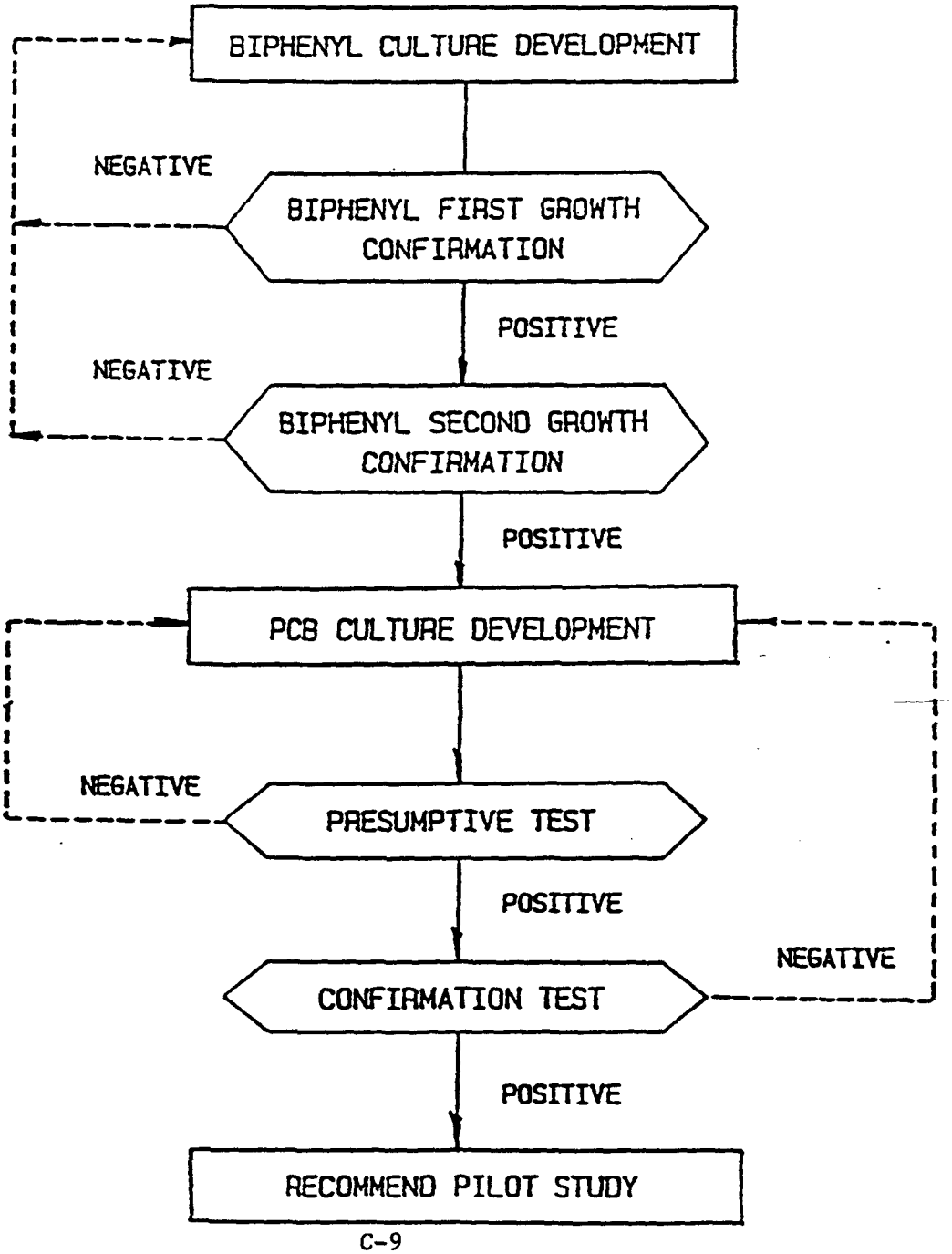
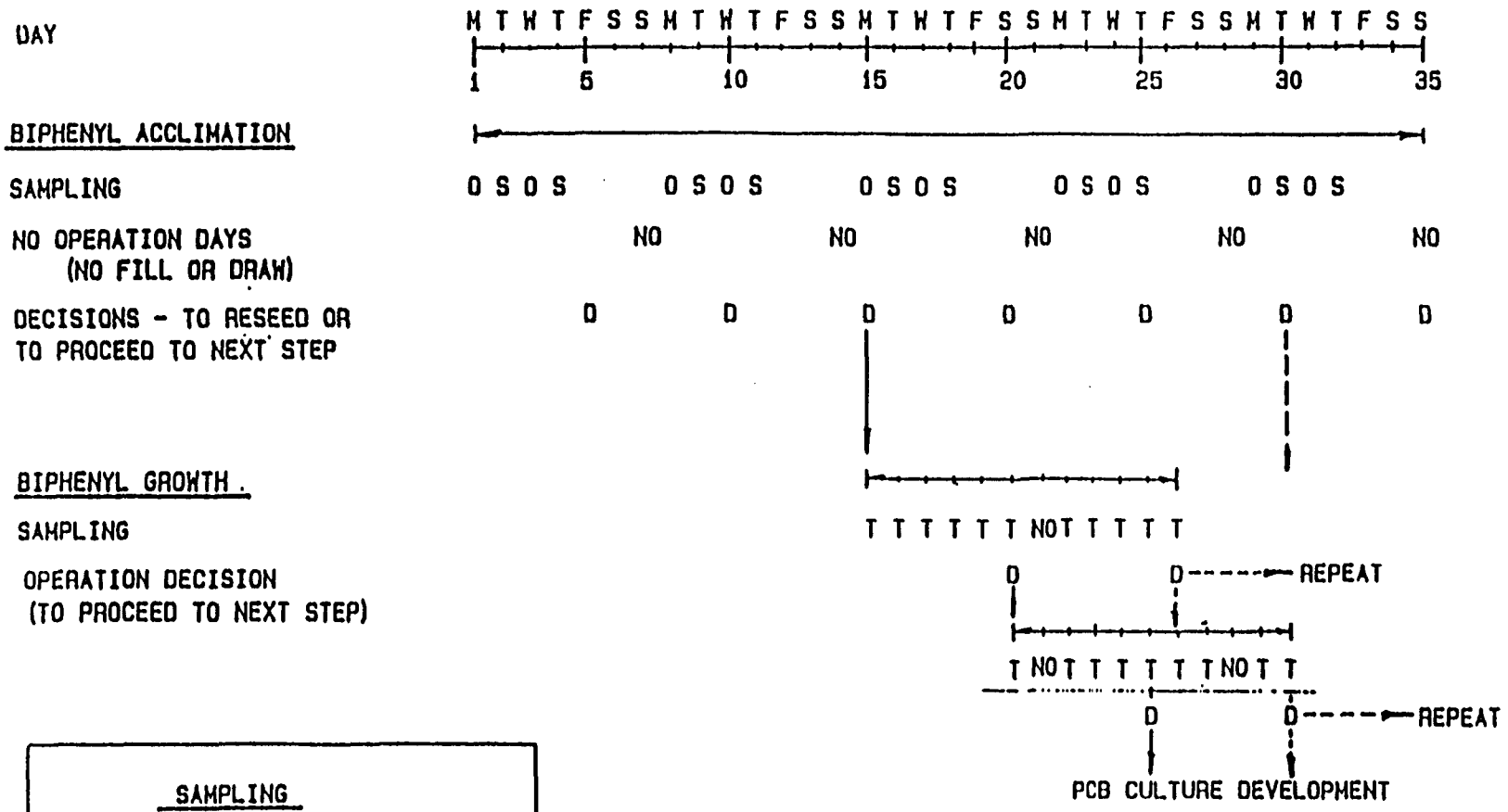


FIGURE 2
DEVELOPMENT OF BIPHENYL CULTURES



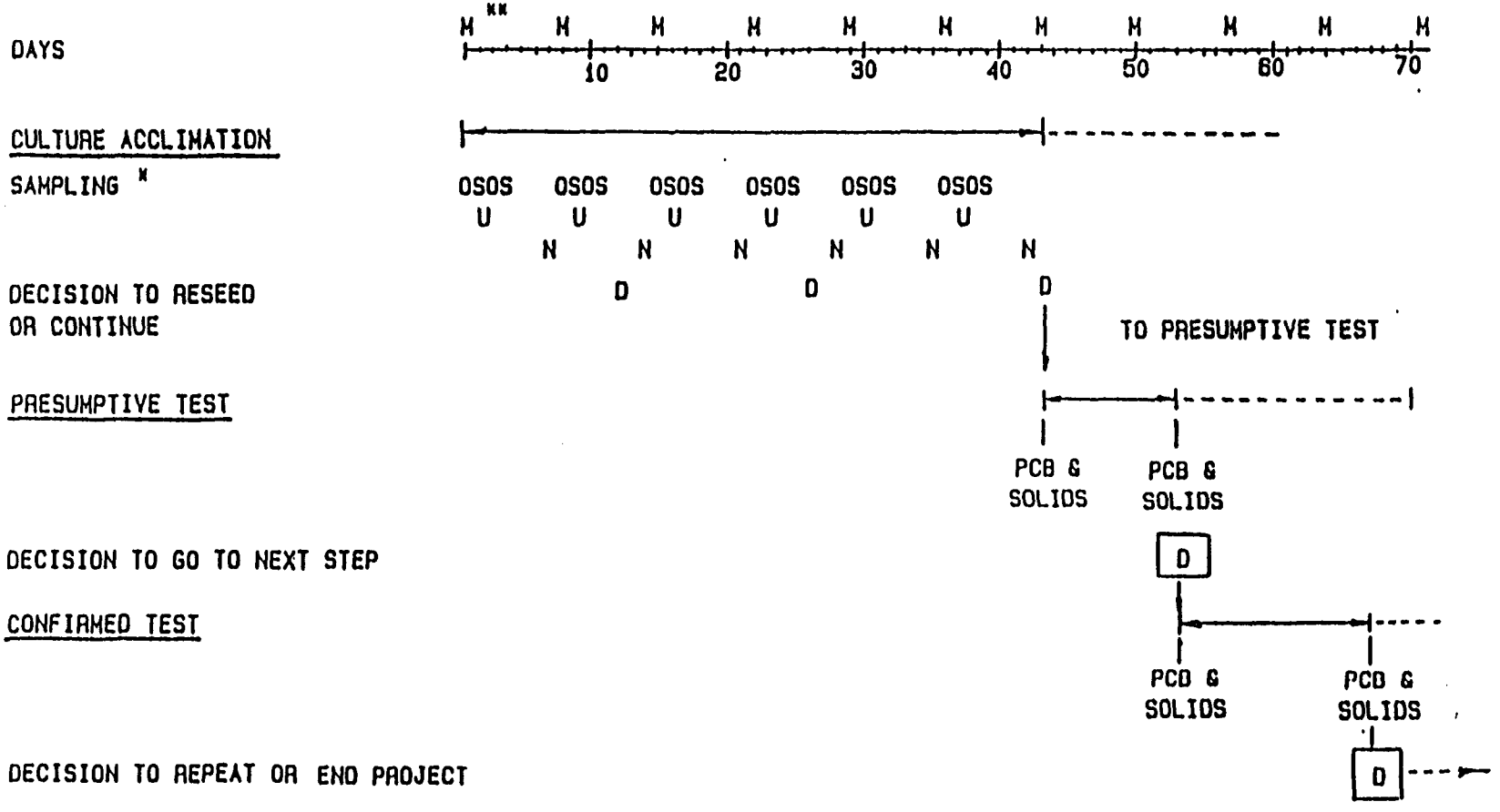
SAMPLING

O = OXYGEN UPTAKE
S = TOTAL & TOTAL VOLATILE SOLIDS
T = TURBIDITY
NO = NO OPERATION

During this phase, PCB contaminated sludge from the harbors at New Bedford, MA, will be used as the primary substrate. Analyses to be performed on a scheduled basis are total solids, total volatile solids, and PCB. Total kjeldahl nitrogen, chemical oxygen demand, and total phosphorous analyses will be done as needed (Figure 3).

Upon completion of the second phase, and analysis of the data, a decision will be made as to the success or failure of the bench scale treatability tests.

FIGURE 3
DEVELOPMENT OF PCB CULTURE



* SAMPLING KEY: O-DISSOLVED OXYGEN; S-TOTAL AND VOLATILE SOLIDS; U-OXYGEN UPTAKE ; N-NO OPERATION

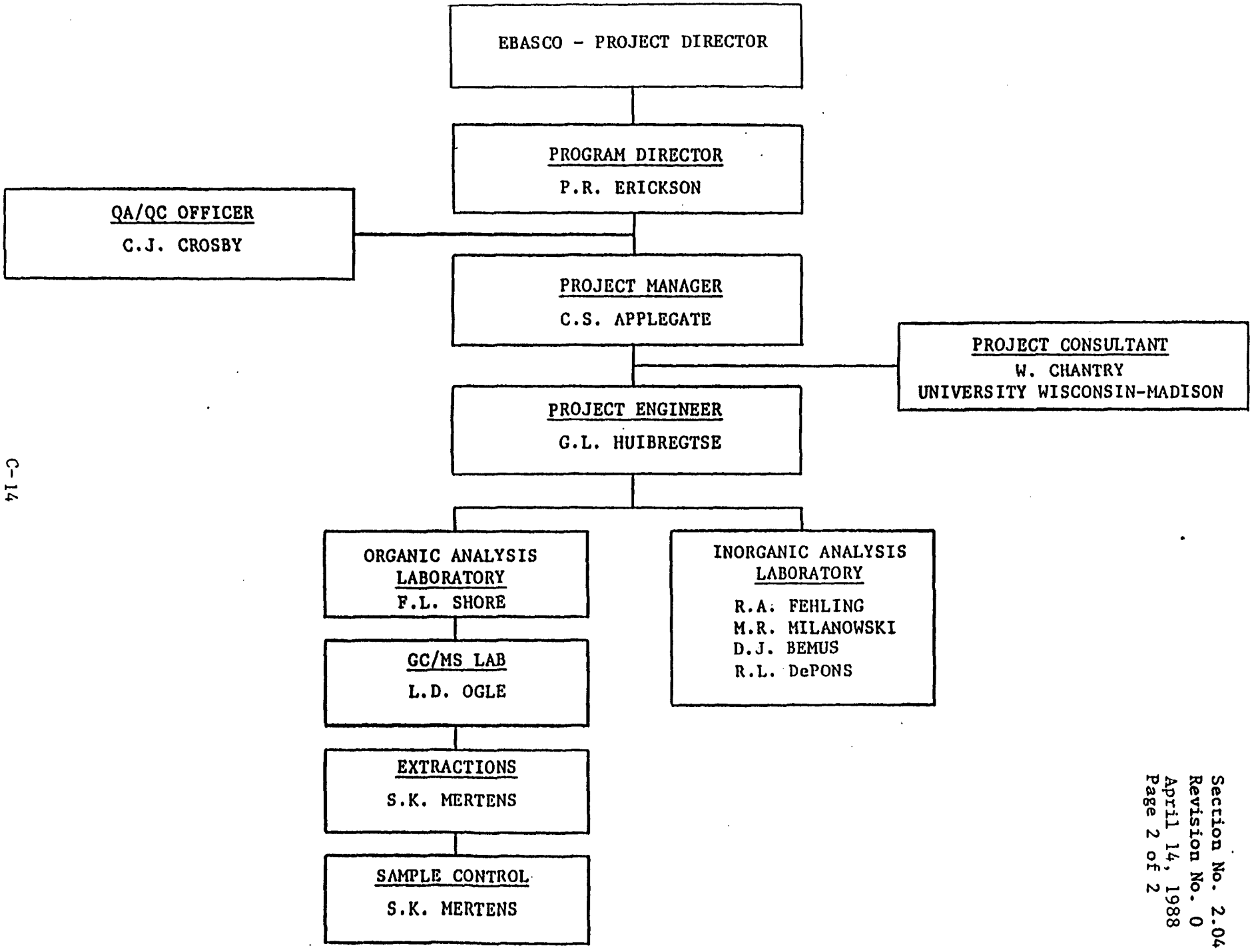
** MONDAY

PROJECT ORGANIZATION AND RESPONSIBILITY

The development of an enriched saltwater microbial community capable of biodegrading PCB's sorbed to harbor sediments will be performed by Radian Corporation - Milwaukee. Radian will plan the test work, install and operate all equipment, obtain samples, and perform conventional analyses. Samples for PCB analysis will be sent to Radian Analytical Services laboratory in Austin, Texas. Additional samples for PCB analysis will also be sent to a laboratory designated by the Sample Management Office of CLP.

A chart showing the project organization is shown in Figure 3. Mr. Paul Erickson of Radian, Milwaukee (MIL) is the Program Director for this project. Mr. Charles Applegate of Radian (MIL) is the Project Manager and has the overall responsibility for his Company's technical performance on this project. Mr. Greg Huibregtse is the Project Engineer and will be responsible for planning and implementing the bench scale tests, evaluating data and preparing reports. He will utilize a staff of engineers and technicians to assist in the bench scale tests and data evaluation. Mr. William Chantry of the University of Wisconsin will be the Project Consultant.

PCB analyses to be conducted by Radian and performed under the direction of Mr. Frank Shore of Radian, Austin and for all other parameters under the direction of Mr. Charles Applegate of Radian, Milwaukee. Mr. Clark Crosby will serve as Quality Assurance Officer and will be responsible for reviewing and approving analytical data for all analyses performed on this project.


EBASCO - PROJECT DIRECTOR
PROGRAM DIRECTOR
P.R. ERICKSON
QA/QC OFFICER
C.J. CROSBY
PROJECT MANAGER
C.S. APPLEGATE
PROJECT CONSULTANT
W. CHANTRY
UNIVERSITY WISCONSIN-MADISON
PROJECT ENGINEER
G.L. HUIBREGTSE
ORGANIC ANALYSIS LABORATORY
F.L. SHORE
INORGANIC ANALYSIS LABORATORY
R.A. FEHLING
M.R. MILANOWSKI
D.J. BEMUS
R.L. DePONS
GC/MS LAB
L.D. OGLE
EXTRACTIONS
S.K. MERTENS
SAMPLE CONTROL
S.K. MERTENS

QUALITY ASSURANCE OBJECTIVE FOR ANALYTICAL MEASUREMENTS

The data quality objectives for precision, accuracy and completeness are listed in Table 1. The values in the precision and accuracy columns are the criteria used to evaluate whether individual duplicate or spiked analysis results are acceptable. Values of relative difference (D_R) and difference between duplicate analyses (D) greater than the listed values and values of recovery (R) outside the listed range indicate unacceptable quality control. Because most of the measured parameters can be rerun if unacceptable data are obtained, it is expected that completion will exceed 95 percent for these parameters.

Table 1. Data Quality Objectives

Parameter	Precision	Accuracy	Completeness
Total Solids	D_R = 15%	Not Measured	>95%
Total Volatile Solids	D_R = 20%	Not Measured	>95%
Turbidity	D_R = 15%	Not Measured	>95%
PCB-USEPA Method 680	D_R = 20%	R = 100 +25 -50	>95%
Salinity	D_R = 15%	Not Measured	>95%
Total COD (TCOD)	D_R = 15%	R = 100+15	>95%
Total Phosphate (TP)	D_R = 15%	R = 100+15	>95%
Ammonia Nitrogen (NH3)	D_R = 15%	R = 100+15	>95%

D_R = relative difference = $\frac{200 \text{ (replicate 1 - replicate 2)}}{\text{replicate 1 + replicate 2}}$

NOTE: Replicate 1 >replicate 2

D = difference between duplicate analysis

R = percent recovery = $\frac{100 \text{ (spike sample - unspiked sample)}}{\text{spike concentration}}$

Completeness = $\frac{100 \text{ (Number of acceptable data values)}}{\text{number of submitted samples}}$

SAMPLING PROCEDURES

Sampling

The sampling schedules will follow the outlines shown in Figures 2 and 3, Section 2.04 of this report. A general description, by phase follows.

PHASE 1 - Biphenyl Culture Development

Acclimation

Growth on the biphenyl, sewage mixture will be monitored by biweekly sampling of the solids from each reactor, and by biweekly oxygen uptakes. Specifically, total solids and total volatile solids will be performed initially and on Tuesdays and Thursdays of each week on the effluent from the reactor. Oxygen uptakes will be performed on Mondays and Wednesdays. Because 300 ml of media is required to perform on O_2 uptake test, the contents of 2 reactors will be combined for the analysis (resulting in 3 analysis per sampling day or 42 O_2 uptake measurements in 7 weeks). The sample will then be redivided into 150 ml fractions and returned to the culture reactors. Mixing pairs of flasks will allow cross seeding of cultures, increasing the opportunity for successful acclimation. Reactor pairs to be mixed will be selected randomly each sampling period.

In addition to these analyses, the total weight of each reactor will be measured prior to each feeding, and water will be added as needed to replace any evaporative losses.

Culture Testing

The turbidity of each of the cultures will be measured 6 of every 7 days of reactor run (Monday thru Saturday). The sample used to determine turbidity will not be harmed and will be returned to the reactors. The weight of the reactor will also be measured on the same schedule to check for evaporative losses.

PHASE 2 - PCB Culture Development

Acclimation

In that during this period, the goal is only to acclimate the microbial culture, no testing of the PCB degradation is necessary. General operation variables will be monitored, including solid levels, O_2 uptakes, and O_2 levels. Both total and total volatile solids will be analyzed on every Tuesday and Thursday, using the effluent from the draw and fill operation.

Dissolved oxygen levels will be measured on Mondays and Wednesdays, oxygen uptake rates will be performed on Wednesdays.

Presumptive Testing

During this project step samples for PCB analysis of the feed to the reactors and effluent from the reactors will be taken for shipment to Jordan Laboratories and for in-house analysis.

To determine the PCB levels and distribution of PCB congeners in the feed to the reactors a 10 day composite sample of the feed to each reactor will be

taken. Specifically, 22 ml of media will be added to the reactors and to a composite sample for each reactor. At the end of the 10 day period the volume of composite feed will be 220 ml. Of this 40 ml (4 x 10ml) will be sent to CLP labs for analysis (sufficient for two homologue tests and two 680 tests). The remaining volume will be used for the in-house analysis for PCB's and for total and total volatile solids. See Table 2 for sample processing schedule.

To determine the corresponding information in the reactor effluents a second set of composites will be taken. The draw from each of the reactors will be composited separately for the 10 day period (220 ml in each of 6 composites). Again 40 ml will be sent to Jordan for analysis from each composite, and the rest used for in-house analysis for PCB's and solids.

Sterilization Test - Overview (See revised Sections 2.06.1 and 2.06.2, Attachment I) Control flasks will be used during the confirmation phase of this project to determine the degree of PCB loss without biological activity. Formaldehyde will be used to sterilize the culture in the control flasks. The level of formaldehyde required will be determined by experimentation at various dosages. Sterilization effectiveness will be determined by total heterotrophic plate count using a marine agar. These tests will be conducted in advance using culture developed in Section 3.1 "Development of PCB Culture", of the Project Work Plan. Approximately 300 ml of culture that has been developed for at least one residence time (14 days) will be used to determine the required formaldehyde dosage. The tests will duplicate conditions of the confirmation test.

TABLE 2 - SAMPLES FOR PCB ANALYSES

	PRESUMPTIVE TEST		CONFIRMATION TEST	
	CLP Samples	Radian Samples	CLP Samples	Radian Samples
Composite Feed			Initial	
	2 high* (608)** 2 high (680) 2 low (608) 2 low (680)	1 high (680) 1 low (680)	8 high live (608) 8 high live (680) 4 low live (608) 4 low live (680) 2 high killed (608) 2 high killed (680) 2 low killed (608) 2 low killed (680)	1 high (680) 1 low (680)
Composite Effluent			Final	
	8 high (608) 8 high (680) 4 low (608) 4 low (680)	2 high (680) 2 low (680)	8 high live (608) 8 high live (680) 4 low live (608) 4 low live (680) 2 high killed (608) 2 high killed (680) 2 low killed (608) 2 low killed (680)	1 high live (680) 1 low live (680) 1 high killed (680) 1 low killed (680)
TOTAL	32 - (duplicate samples from each source for each method)	6	64 - (duplicate samples from each source for each method)	6

* - high and low = PCB concentrations in sediments
** - (680) & (608) - EPA Methods

During the confirmation test, formaldehyde concentration will be monitored using the Hantzsch Reaction Method. Additional formaldehyde will be added on a daily basis for the first 6 days to maintain the concentration determined to be effective in the pre-test. At the end of 6 days, a plate count will be made to confirm sterilization. Formaldehyde dosage will continue at levels determined in the first 6 days of monitoring.

Test Procedures: Formaldehyde Sterilization

After at least one retention time (14 days) one of seven shake flasks containing about 300 ml of low level PCB culture will be taken out of service for sterilization tests. 50 ml portions will be placed in 6 flasks on an orbital shaker. Formaldehyde will be dosed as a 30% solution in the following concentrations base on weight percent formaldehyde.

FLASK#	FORMALDEHYDE CONCENTRATION
	%
1	0
2	1
3	3
4	5
5	5
6	10

Flask #1 is an undosed control and flasks 4 and 5 are duplicates.

The flasks will be incubated at 20°C while shaking for 14 days. At the end of 14 days numbers of culturable, aerobic and facultative anaerobic heterotropic bacteria will be determined by plating on marine agar. The procedure used will substantially follow "Standard Methods for Examination of Water and Wastewater", American Public Health Association, 16th Edition; Method 907. However, Bacto Marine Agar 2216 (Difco Laboratories, Detroit, Michigan) will be substituted for the specified agar media. The media will be prepared in accordance with Difco's recommendations. A copy of the appropriate section from the Difco Manual Can be found in Attachment I.

The Pour Plate Method (SM 907A) will be used for preparing the plates. Four dilutions will be prepared in triplicate in the range of 10^{-1}-10^{-5}. On sterilized samples, one set will be prepared with no dilution.

Formaldehyde Determination in Control Culture

During the confirmation tests, formaldehyde concentration will be monitored during the first 6 days and formaldehyde will be added to maintain the dosage determined in the pre-test. The procedure will be by the Hantzsch Reaction Method given in Formaldehyde, Walker, 3rd Edition p. 472.

A copy of the procedure can be found in Attachment I. The procedure is based on the reaction of formaldehyde with acetylacetone and an ammonium salt to give diacetyldihydrolutidine.

On day 6 of the PCB confirmation tests, heterotrophic plate counts, using the previously outlined procedure will be performed on the two control flasks to confirm sterilization efficiency (see section titled "Test Procedures: Formaldehyde Sterilization").

Confirmation of PCB Biodegradation

These reactors will be sampled for PCB's and solids levels at the beginning and end of the 14 day period. For sampling of the initial conditions, 8-100 ml samples of media split from the media mixed for each reactor will be taken. Of this, 8-40 ml samples will be sent to Jordan for PCB analysis, the rest used for in-house PCB and solids analysis. Two in-house PCB analyses, one on a high PCB sediment media, and one on the lower PCB sediment media will be performed. Total and total volatile samples will be determined on each of the 8 reactor samples.

For sampling of the reactor contents at the end of the 14 day run, the entire contents of the reactors will be available for analysis. Again, 8 samples of 40 ml each will be sent to CLP labs for PCB analysis. Two in-house PCB analyses will be performed, and 8 sets of solids analyses.

Sample Bottles

New polyethylene sample bottles with polyethylene foam lined plastic caps will be used for non-PCB samples. Prior to use, the bottles will be washed with detergent, rinsed with tap water and demineralized water, and dried. Cleaned bottles will be capped until used.

Amber glass bottles with Teflon R lined caps will be used for PCB analyses (30 ml volume). The bottles will be purchased precleaned from I-Chem Research. The following method will be used by I-Chem.

1. Wash containers, closures and teflon liners in hot tap water with laboratory grade nonphosphate detergent.
2. Rinse three times with tap water.
3. Rinse one time with 1:1 nitric acid.
4. Rinse three times with ASTM Type 1 deionized water.
5. Rinse one time with pesticide grade methylene chloride.
6. Oven dry.
7. Remove containers, closures, and teflon liners from oven.
8. Place teflon liners closures and place closures on containers. Attendant to wear gloves and containers to not be removed from preparation room until sealed.

Sample Preservation and Handling

Sample preservation, if necessary for the parameters to be analyzed, will be performed as soon as possible after collection by the individuals collecting the samples. The following preservation methods will be used:

Parameters	Preservation
Volatile Total Solids	Keep at $4^{o}C$
TCOD, total phosphate, TKN	Acidify to pH<2 with H_2SO_4 keep at $4^{o}C$
Specific Organic Compounds (PCB)	Keep at $4^{o}C$ - 7 days before extraction or 40 days after, pH 5-9
Salinity, turbidity	Keep at $4^{o}C$

Samples will be maintained at 4°C or kept in contact with ice from time of sample preservation until they are discarded. Composite samples will be kept in contact with ice during entire compositing period.

Sample Identification

All sample identification and custody is organized around the Sample and Analysis Management System (SAM). SAM is a computer hardware and software system specifically designed for tracking and handling the large amount of information required for the efficient management of an analytical chemistry laboratory. The identification of each sample will also be entered into the field sampling log. The information to be entered includes:

Sample ID Number
Ebasco Project No.
Sample Description
Sample Location
Sample Type: Composite or grab
    If composite, record start and end time and date of compositing period.
    If grab, record time and date of sample.
Initials of sampler.

Measurements performed in-situ (dissolved oxygen) and samples taken for settling tests will not require sample ID numbers. The information from these measurements will be entered directly on the appropriate operation data sheets along with time and date of measurement, sample description, location and identity of the person taking the measurement.

Sample Holding Time

The sample holding times listed below will be used. These are the maximum times between sample collection (or end of compositing period) and the start of analyses for properly preserved samples kept at 4°C or less.

Parameter	Maximum Holding Time
TS, TVS	7 days
TCOD	28 days
TKN	28 days
Phosphates	28 days
Salinity	1 hour-unsealed Indefinitely-sealed
Turbidity	48 hours
PCB	7 days till extraction 40 days after extraction

SAMPLE CUSTODY

The identification of samples and the information to be entered in the project log were described in Section 2.06. Each sample bottle will be labeled at the time of sample collection with project number, sample description, preservative (if any), sample I.D. number, and sample type.

Sample chain-of-custody sheets will be prepared for samples shipped from Radian-Milwaukee to Radian-Austin and to CLP labs. These chain-of-custody sheets will include the sample ID number, date and time of sampling, preservatives added (if any) and the analyses or tests to be performed. A brief description of each sample will also be included. The custody sheets will also include the identity of the person packaging the samples, the transportation method used and date of shipment.

The chain-of-custody sheet will be completed in quadruplicate. Two copies will accompany the sample to the laboratory, one will be kept by the Project Engineer, and one will be sent to Jordan for the project file. Upon receipt of samples, the receiving laboratory will be requested to enter the time and date of arrival, the identities of the carrier and the person receiving the samples and the condition of the samples on the chain-of-custody sheet. The sheet will be returned to the Project Engineer. The receiving laboratories will then enter the samples in their laboratory sample control system and be responsible for proper storage and holding time.

CALIBRATION PROCEDURES AND FREQUENCY

Calibration procedures for the anaylses to be performed will follow acceptable laboratory practice for State of Wisconsin Certified Laboratories. Attachment I summarizes calibration procedures, frequencies, and acceptance criteria for the analytical methods defined by Chapter NR149 of the Wisconsin Administrative Code.

PCB compounds measured by EPA Method 680 will use instruments calibrated by procedures as specified by the method.

ANALYTICAL PROCEDURES

The EPA analytical procedures that will be used for analysis of parameters common to water analysis are described in detail in Methods for Chemical analysis of Water and Wastes, EPA-600/4-79-020, revised March, 1983.

PCB analyses will follow specific analytical procedures as outlined in EPA Method 680, "Determination of Pesticides and PCB in Water and Soil/Sediment by Gas Chromatography/Mass Spectroscopy".

DATA REDUCTION, VALIDATION AND REPORTING

Specific procedures for calculating concentrations of measured parameters from analytical measurements are described in the designated analytical procedures listed in Section 2.09. Raw data reduction is the responsibility of the analysts who generate the data. All data and calculation reductions will be reviewed by the QA Officer. Original analytical data will be recorded in laboratory notebooks.

Results of measured parmeters will be entered into a computerized data tracking and reporting system. Reports, final and interim, will present the analytical results and assess the supporting quality assurance and quality control data. All reports will be signed by the Radian Project Manager.

INTERNAL QUALITY CONTROL CHECKS

The quality control checks used for analytical parameters common to water analysis will follow guidelines as specified in State of Wisconsin Administrative Code NR149 (Attachment 1). PCB analyses will follow quality control checks specified in EPA Method 680. Additionally, quality control checks specified in the Radian-Milwaukee Quality Assurance Program Plan will be followed.

PERFORMANCE AND SYSTEM AUDITS

Radian-Milwaukee participates in the State of Wisconsin Laboratory Certification Program which requires detailed analysis of reference samples (up to 3 per year). Renewal of certification is completed on a yearly basis. Radian-Milwaukee is a State of Wisconsin certified laboratory.

Radian-Milwaukee also uses blind reference samples from Environmental Resource Associates (ERA) to assess laboratory performance on a quarterly basis. The results of the blind QA samples are used to identify and correct problem analyses.

Radian-Austin participates in EPA performance evaluations for the WP and WS series as well as the CLP organic audits. In-house systems audits of the analytical programs are also routinely conducted and cover equipment calibration, records, and sampling activities.

PREVENTIVE MAINTENANCE

The preventive maintenance program used by Radian is designed to minimize the downtime of analytical equipment. Routine preventive maintenance is performed on all instruments at Radian. Maintenance is based on manufacturers' recommendations and is performed on a regular schedule. All aspects of routine and nonroutine instrument maintenance are recorded in logbooks. Standard operating procedures for the laboratories incorporate instrument and equipment maintenance into the overall laboratory quality assurance program.

SPECIFIC ROUTINE PROCEDURES USED TO ASSESS DATA PRECISION, ACCURACY AND COMPLETENESS

The specific procedures to be used to calculate precision and accuracy are listed below:

Difference between duplicates -

$D = X_1 - X_2$ where

D = difference between duplicates

X_1 and X_2 are duplicate measurements

Standard deviation -

$$S = \sqrt{\frac{(X_i - X)^2}{n-1}} \quad \text{where}$$

S = standard deviation

X_i = value of replicate i

X = average value of all replicates

n = number of replicates

Σ = indicates the sum of the term $(X_i - X)^2$ for each replicate from i = 1 to n

Coefficient of variation (%) -

$$C_v = \frac{100\ S}{X} \quad \text{where}$$

C_v = coefficient of variation (%)

S = standard deviation

X = average value

Relative difference (%) -

$$D_R = \frac{200\ (X_1 - X_2)}{X_1 + X_2} \quad \text{where}$$

D_R = relative difference (%)

X_1 = value of replicate

X_2 = value of second replicate

Recovery (%) -

$$R = \frac{100\ (X_s - X_u)}{X_k}$$

R = percent recovery

X_s = concentration in spiked sample

X_u = concentration in unspiked sample

X_k = concentration added by spiking

CORRECTIVE ACTION

The internal quality control checks for the analytical procedures are listed in Section 2.11. When the control checks indicate a problem, the initial corrective action is taken by the analyst. Examples of corrective action are:

1. Check instrument and equipment operation parameters and rerun.

2. Check quality of standards, spiking materials and/or reagents. Replace suspect chemicals and rerun.

3. Check cleanliness of analytical equipment and materials. Clean or replace suspected parts.

If these actions do not correct the problem, the analysts supervisor will be consulted for further action which includes: 1) determine the cause of the problem and 2) determine appropriate corrective action. Some causes of undesirable levels of precision or accuracy are:

1. Interfering substances

2. Insufficient sample preparation

3. Improper sample handling

4. Poor analytical technique

Once the cause is determined, appropriate corrective actions can be performed. Corrective action resulting from interferences, insufficient sample preparation or improper sample handling must be approved by the Laboratory Manager (or his designated representative) and reported to the Quality Assurance Officer. All corrective actions must be recorded in the appropriate quality control log.

When the problem has been corrected, sample analyses will resume. Samples analyzed while the analytical procedure was not in control will be rerun if sample storage and holding time criteria are met.

QUALITY ASSURANCE REPORTS TO MANAGEMENT

Results of project data precision and accuracy will be submitted to the project Quality Assurance Officer with the analytical data. Results of performance audits will also be submitted when completed. Reports of corrective actions that may have an impact on data evaluation will be forwarded to the Quality Assurance Officer by the Laboratory Manager as required in Section 2.15.

Upon completion of a data validation review or system performance audit, the QA Officer will submit a report to the Project Manager identifying the approved data, the data that was rejected and the reasons for the rejection.

A T T A C H M E N T I

STERILIZATION TEST

HANTZSCH FORMALDEHYDE TEST

2.06 Sterilization Test - Overview

Control flasks will be used during the conformation phase of this project to determine the degree of PCB loss without biological activity. Formaldehyde will be used to sterilize the culture in the control flasks. The level of formaldehyde required will be determined by experimentation at various dosages. Sterilization effectiveness will be determined by total heterotrophic plate count using a marine agar. These tests will be conducted in advance using culture developed in Section 3.1 "Development of PCB Culture", of the Project Work Plan. Approximately 300 ml of culture that has been developed for at least one residence time (14 days) will be used to determine the required formaldehyde dosage. The tests will duplicate conditions of the confirmation test. Detailed procedures are provided in Section 2.06.1.

During the confirmation test, formaldehyde concentration will be monitored using the Hantzsch Reaction Method. Additional formaldehyde will be added on a daily basis for the first 6 days to maintain the concentration determined to be effective in the pre-test. At the end of 6 days, a plate count will be made to confirm sterilization. Formaldehyde dosage will continue at levels determined in the first 6 days of monitoring. Detailed procedures for formaldehyde analysis are given in Section 2.06.2.

2.06.1 Test Procedures: Formaldehyde Sterilization

After at least one retention time (14 days) one of seven shake flasks containing about 300 ml of low level PCB culture will be taken out of service for sterilization tests. 50 ml portions will be placed in 6 flasks on an orbital shaker. Formaldehyde will be dosed as a 30% solution in the following concentrations base on weight percent formaldehyde.

FLASK#	FORMALDEHYDE CONCENTRATION
	%
1	0
2	1
3	3
4	5
5	5
6	10

Flask #1 is an undosed control and flasks 4 and 5 are duplicates.

The flasks will be incubated at 20°C while shaking for 14 days. At the end of 14 days numbers of culturable, aerobic and facultative anaerobic heterotropic bacteria will be determined by plating on marine agar. The procedure used will substantially follow "Standard Methods for Examination of Water and Wastewater", American Public Health Association, 16th Edition; Method 907. However, Bacto Marine Agar 2216 (Difco Laboratories, Detroit, Michigan) will be substituted for the specified agar media. The media will be prepared in accordance with Difco's recommendations. A copy of the appropriate section from the Difco Manual is attached.

The Pour Plate Method (SM 907A) will be used for preparing the plates. Four dilutions will be prepared in triplicate in the range of 10^{-1}-10^{-5}. On sterilized samples, one set will be prepared with no dilution.

2.06.2 Formaldehyde Determination in Control Culture

During the confirmation tests, formaldehyde concentration will be monitored during th first 6 days and formaldehyde will be added to maintain the dosage determined in the pre-test. The procedure will be by the Hantzsch Reaction Method given in Formaldehyde, Walker, 3rd Edition p. 472.

A copy of the procedure is attached. The procedure is based on the reaction of formaldehyde with acetylacetone and an ammonium salt to give diacetyldihydrolutidine.

On day 6 of the PCB confirmation tests, heterotropic plate counts, using the procedures in Section 2.06.1 will be performed on the two control flasks to confirm sterilization efficiency.

HANTSCH FORMALDEHYDE TEST

Description

This test is to determine the formaldehyde concentration in water samples.

Apparatus

1. Bausch & Lomb Spectronic 100 20 spectrophotometer.
2. A 55°C water bath.
3. 25 ml. Kimax graduated test tubes and corks.
4. 2-10 ml. pipettes.

Reagents

Acetylacetone Reagent Reference: Formaldehyde, Walker. 3rd Edition, p. 472

1. 150 grams of ammonium acetate.
2. 3 mls. of acetic acid (glacial).
3. 2 mls. of 2,4 pentanedione (redistilled acetylacetone).

Place the above reagents in a one liter volumetric flask and fill to mark with distilled water; keep stoppered. This solution should be replaced every three months.

Procedure

1. Set the Spectronic 100 20 at a wavelength of 420 nm.
2. Pipette 10 mls. of distilled water into 25 ml. test tube for a blank. Pipette 10 mls. of sample into another 25 ml. test tube.
3. Pipette 10 mls. of acetylacetone solution into each of the 25 ml. test tubes. Cork each tube and place in 55°C water bath for 5 minutes or 35°C bath for 40 minutes. Immediately cool under cold water tap. Transfer samples to 1/2 inch cells and read % Transmittance immediately.
4. To use accompanying chart read % Transmittance to determine the µg/ml (ppm) of formaldehyde in the sample.

Text book

FORMALDEHYDE, WALKER · 3rd Edition

Methanol and ethanol do not interfere with this procedure, but higher alcohols hinder color formation. Acetaldehyde, acrolein and beta-hydroxypropionaldehyde interfere, but formaldehyde can be determined in the presence of 100 times as much acetaldehyde if 300 mg of reagent is employed in place of the customary 50 mg. Benzaldehyde does not interfere. Acetone causes the color to fade when the solution is diluted with water, but this can be avoided by diluting to 50 ml with 18*N* sulfuric acid. Methyl ethyl ketone and diacetone alcohol interfere.

Bricker and Johnson [13] note that the light absorbance of the chromotropic acid analysis mixture is proportional to temperature. Lee [36] suggests the use of concentrated sulfuric acid as a diluent in the analysis to avoid temperature effects whereas Morath and Woods [42] report that 5*N* sulfuric acid reduces the temperature effect and prefer it for reasons of safety.

West and Sen [57] describe a modified chromotropic acid procedure which is stated to give a stable color that obeys Beer's law over a wide concentration. This procedure is also reported to diminish the effect of acrolein which is the only impurity stated to have a serious action on the results. To accomplish this, West and Sen [57] prepare a 1 per cent solution of chromotropic acid in concentrated sulfuric acid which is then filtered through a sintered glass crucible. In carrying out the test, 1 ml of the formaldehyde solution is added to 1 ml of the chromotropic acid solution to which concentrated sulfuric acid is then added in sufficient excess to insure a mixture containing at least 86 per cent of this acid. The mixture is then cooled and adjusted to the desired volume and its transmittance determined for 570 millimicron light. The analysis sample employed in this procedure may contain 0.05 to 2.0 micrograms of formaldehyde per ml. No external heating other than the heat of dilution is required. Altshuller, Miller and Sleva [4] have checked this method and recommend it.

Hantzsch Reaction Method

The Hantzsch reaction method as developed by Nash [44] is unique in that it may be carried out under mild conditions so that it can be used in the presence of living material. The procedure is based on the reaction of formaldehyde with acetylacetone and an ammonium salt to give diacetyldihydrolutidine. This lutidine derivative is yellow and under preferred conditions its molecular extinction in terms of formaldehyde has a maximum of 8000 at 412 millimicrons. The procedure for carrying out the analysis as recommended by Nash [44] is as follows:

Procedure. *Preparation of reagent:* Dissolve 150 g of ammonium acetate, 3 ml of acetic acid and 2 ml of redistilled acetylacetone in one liter of water. This gives a solution which is approximately 2 molal in ammonium acetate, 0.05 molal in acetic acid and 0.02 molal in acetylacetone.

Test: The reagent is mixed with the same volume of sample solution containing not over 8 micrograms of formaldehyde per milliliter. The time required for optimum color development varies with temperature. At 20, 37 and 58°C respectively, the time intervals should be 5 hours, 40 minutes and 5 minutes. The color should be measured in a photoelectric colorimeter with a deep, blue filter at about 412 millimicrons.

The reagent used in this procedure has a control absorption value of about 1 per cent which does not increase for over one week if the reagent is kept from exposure to formaldehyde vapors. The calibration absorption curve does not give a straight line when the logarithm of the intensity is plotted against formaldehyde concentration. However, according to Nash [44], this is not due to a departure from Beer's law but is a result of the inefficiency of the blue light filters.

The Hantsch reaction method is highly specific under the mild conditions required if the reaction time is kept to a minimum. Acetaldehyde produces diacetyldihydrocollidine which has a maximum absorption at 388 millimicrons. However, the formation rate is lower so that little interference is noted when approximately equimolar portions of formaldehyde and acetaldehyde are present in the sample. The method is not satisfactory in the presence of large amounts of acetaldehyde. Nash [44] reports that acetone, chloral, furfural and glucose do not interfere. Pure ninhydrin is also stated to have no appreciable effect, this material being equivalent to about 0.005 molecular proportions of formaldehyde.

Belman [45] makes use of the fluorescence of the diacetyldihydrolutidine produced in the Hantzsch reaction for determining trace concentrations of formaldehyde. For this purpose, a fluorimeter equipped with a primary 405-millimicron interference filter is employed. A reaction temperature of 37°C is preferred and measurements are made at room temperature. The fluorescence curve is linear for 0.005 to 0.4 microgram per milliliter formaldehyde but deviates somewhat in the 0.4 to 1 microgram range. Higher concentrations are readily measured by colorimetry. Belman has found the fluorimetric method of special value for determining low formaldehyde concentrations in the presence of nucleic acids and other biological materials.

Miscellaneous Color Tests

A wide range of color tests of varying degrees of sensitivity and specificity have been reported for the detection and estimation of formaldehyde in addition to the three methods previously described. West and

Sen [57] investigated a number o
work on the chromotropic acid
acid and 2,7-dihydroxynaphth
workers [50] studied spot tests
and recommend a 1 per cent
sulfuric acid which develops
dehyde on steaming. These in
gives a blue color with forma
yellow color with pyruvaldehy

Schryver's test [32] involves t
per cent potassium ferricyanid
cent phenylhydrazine hydroch
chloric acid to 10 ml of the s
ent, a fuchsin-red color develo
is reported to give a distinct t
sunlight in the presence of moi
report a colorimetric method in
are oxidized in an alkaline med
absorption at 512 millimicron

Alkaloids have long been us
ported to have a high degree
a purple coloration with morph
acid [10,31]. A convenient proced
morphine hydrochloride to a
form an underlayer of sulfuric
when the sample contains
phine [27,60] give similar reaction

Fulton [25] found that the addi
cent solution of ferric sulfate c
ml water, increases the sensitiv
Thus, apomorphine, codeine,
ppm, whereas papaverine, clai
10 ppm formaldehyde.

Voisenet [55] reports that an e
0.1 ppm formaldehyde can b
nitrite. One drop of the unkno
tion containing 0.05 g albumin
of 0.5 ml of 3.6 per cent potas
chloric acid. A rose color for
standing for 5 minutes when

Tollen's ammoniacal silver

REFERENCES
1. J. Bact., 50:201, 1945.
2. Zentr. Bakt., I Abt. Orig., 149:122, 1942.
3. Am. J. Vet. Res., 8:173, 1947.
4. Am. J. Vet. Res., 8:275, 1947.

PACKAGING

Bacto Mannitol Salt Agar	1 lb (454 g)	0306-01-0
	1/4 lb (114 g)	0306-02-9
	5 lb (2.27 kg)	0306-05-6
	10 kilo	0306-08-3

BACTO MANNITOL SALT BROTH

INTENDED USE
Bacto Mannitol Salt Broth is a modified formulation for the detection and isolation of pathogenic staphylococci from foods.

PRINCIPLES
Bacto Mannitol Salt Broth contains only mannitol as a fermentable carbohydrate, the indicator phenol red for ease of detection and an unusually high concentration of salt to inhibit undesired organisms.

FORMULA

BACTO MANNITOL SALT BROTH
DEHYDRATED

Ingredients per liter

Bacto Tryptone	17 g
Pancreatic Digest Casein	
Bacto Soytone	3 g
Papaic Digest Soy Bean Meal	
Bacto Mannitol	2.5 g
Sodium Chloride	100 g
Dipotassium Phosphate	2.5 g
Phenol Red	0.025 g

Final pH 7.3 ± 0.2 at 25°C.

One pound will make 3.63 liters of final medium.

METHOD OF PREPARATION
1. Suspend 125 g in 1 liter distilled or deionized water. Heat to boiling to dissolve completely.
2. Dispense into tubes or flasks as desired.
3. Sterilize in the autoclave for 15 minutes at 15 lbs pressure (121°C).
4. Allow to cool to at least 37°C and inoculate with specimen or culture.
5. Incubate at 35°C for 18 – 48 hours. Seventy-two hours may be required for some organisms.

STORAGE

Bacto Mannitol Salt Broth	Below 30°C
Prepared medium	15 – 30°C

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QUALITY CONTROL

	Identity Specifications
Dehydrated powder:	pink, homogeneous, free-flowing
Reaction of 12.5% solution:	pH 7.3 ± 0.2 at 25°C
Prepared medium:	red, clear to very slightly opalescent

Typical Cultural Response in Bacto Mannitol Salt Broth After 18 – 48 Hours at 35°C

Organism	Growth	Acid
Escherichia coli ATCC® 25922	inhibited	–
Proteus vulgaris ATCC® 13315	inhibited	–
Staphylococcus aureus ATCC® 25923	good to excellent	+
Staphylococcus epidermidis ATCC® 12228	good to excellent	slight + or –

+ = positive, yellow
– = negative, no change, red

PACKAGING

Bacto Mannitol Salt Broth	1 lb (454 g)	0926-01-0

BACTO MARINE AGAR 2216
BACTO MARINE BROTH 2216

INTENDED USE
Bacto Marine Agar 2216 and Bacto Marine Broth 2216 are recommended for culturing heterotropic marine bacteria. The agar medium can also be used to isolate and enumerate these organisms.

HISTORY/PRINCIPLES
Bacto Marine Agar 2216 and Bacto Marine Broth 2216, prepared according to the formula of ZoBell[1,2] contain all of the nutrients necessary for the growth of marine bacteria. Besides including minerals which nearly duplicate the major mineral composition of sea water,[3] it contains peptone and yeast extract which were reported by Jones[4] to be the best source of nutrients for marine bacteria in general.

Marine bacteria are present in nutrient sea water by the millions per ml and are essential to the life cycle of all marine flora and fauna.[5] Because of their increasing importance to the food industry for the conservation of marine life, the enumeration and activity of marine bacteria are important.

In the use of Bacto Marine Agar 2216, the conventional pour plate and spread plate techniques of enumeration are used. For the pour plate technique the agar must be carefully cooled to 42°C before inoculation because of the thermo-sensitive nature of most marine bacteria. In the spread plate technique the agar is poured while hot and allowed to cool and solidify before inoculation. This latter method was reported by Buck and Cleverdon[6] to give higher counts than the pour plate method because of the increased growth of the thermo-sensitive bacteria.

FORMULAE

BACTO MARINE AGAR 2216
DEHYDRATED

Ingredients per liter

Bacto Peptone	5 g	Potassium Bromide	0.09 g
Bacto Yeast Extract	1 g	Strontium Chloride	0.034 g
Ferric Citrate	0.1 g	Boric Acid	0.022 g
Sodium Chloride	19.45 g	Sodium Silicate	0.004 g
Magnesium Chloride	8.8 g	Sodium Fluoride	0.0024 g
Sodium Sulfate	3.24 g	Ammonium Nitrate	0.0016 g
Calcium Chloride	1.8 g	Disodium Phosphate	0.008 g
Potassium Chloride	0.55 g	Bacto Agar	15 g
Sodium Bicarbonate	0.16 g		

Final pH 7.6 ± 0.2 at 25°C.

One pound will make 8.25 liters of medium.
Use 55.1 g per liter.

BACTO MARINE BROTH 2216
DEHYDRATED

Ingredients per liter

Bacto Peptone	5 g	Sodium Bicarbonate	0.16 g
Bacto Yeast Extract	1 g	Potassium Bromide	0.09 g
Ferric Citrate	0.1 g	Strontium Chloride	0.034 g
Sodium Chloride	19.45 g	Boric Acid	0.022 g
Magnesium Chloride Dried	5.9 g	Sodium Silicate	0.004 g
Sodium Sulfate	3.24 g	Sodium Fluoride	0.0024 g
Calcium Chloride	1.8 g	Ammonium Nitrate	0.0016 g
Potassium Chloride	0.55 g	Disodium Phosphate	0.008 g

Final pH 7.6 ± 0.2 at 25°C.

One pound will make 12.13 liters of medium.
Use 37.4 g per liter.

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METHOD OF PREPARATION
1. Suspend appropriate amount in 1 liter cold distilled or deionized water. Heat to boiling to dissolve completely.
2. Distribute into tubes or flasks as desired.
3. Sterilize in the autoclave for 15 minutes at 15 lbs pressure (121°C).

STORAGE
Bacto Marine Media 2216	Below 30°C
Prepared media	15 – 30°C

QUALITY CONTROL
Identity Specifications

Dehydrated powder:	light beige with a few dark particles, free-flowing
Reaction of appropriate solution:	pH 7.6 ± 0.2 at 25°C
Prepared medium:	light amber, clear to slightly opalescent, may have a slight precipitate

Typical Cultural Response in/on Bacto Marine Media 2216
After 2 – 3 Days (longer, if required) at 20°C

Organism	Growth
Vibrio fischeri	good to excellent
Vibrio harveyi	good to excellent

REFERENCES

1. J. Marine Research, 4:62, 1941.
2. Personal Communication.
3. J. Marine Research, 2:134, 1939.
4. Biol. Proc., Pg. 36 (A28), 1958.
5. Ecology, 40:712, 1959.
6. Limnology and Oceanography, 5:78, 1960.

PACKAGING
Bacto Marine Agar 2216	1 lb (454 g)	0979-01-6
Bacto Marine Broth 2216	1 lb (454 g)	0791-01-2

BACTO McCLUNG TOABE AGAR BASE

INTENDED USE
Bacto McClung Toabe Agar Base is recommended for the detection and isolation of *Clostridium perfringens* in foods.

HISTORY/PRINCIPLES
Bacto McClung Toabe Agar Base, prepared according to the formulation of McClung and Toabe,[1,2,3] differentiates various species of *Clostridium* on the basis of their lecithinase production. Lecithinase lyses egg yolk lecithin, producing an opaque precipitate in the agar surrounding the slightly raised colonies.

FORMULA
BACTO McCLUNG TOABE AGAR BASE
DEHYDRATED

Ingredients per liter

Proteose Peptone, Difco	40 g
Bacto Dextrose	2 g
Sodium Phosphate Dibasic	5 g
Potassium Phosphate Monobasic	1 g
Sodium Chloride	2 g
Magnesium Sulfate	0.1 g
Bacto Agar	25 g

Final pH 7.6 ± 0.2 at 25°C.

One pound will make 6 liters of medium.

METHOD OF PREPARATION
1. Suspend 75 g in 1 liter distilled or deionized water. Heat to boiling to dissolve completely.
2. Dispense into flasks in 90 ml amounts.
3. Sterilize in the autoclave for 20 minutes at 15 lbs pressure (121°C).
4. Allow to cool to 50 – 55°C and add 10 ml Bacto Egg Yolk Enrichment 50% to each 90 ml of medium.
5. Mix thoroughly and pour into sterile Petri dishes in approximately 15 ml amounts.

PROCEDURE
1. Place approximately 25 g of food sample to be tested in each of 2 tubes containing 25 ml Bacto Fluid Thioglycollate Medium with fermentation vials (Durham tubes).
2. Incubate inoculated tubes at 46°C for 4 – 6 hours. Observe for growth and gas formation.

A T T A C H M E N T I I

STATE OF WISCONSIN
DEPARTMENT OF NATURAL RESOURCES

LABORATORY CERTIFICATION CODE NR149

NR 149.14 Quality control. (1) Each laboratory shall maintain a quality control program. The quality control program shall be documented and such documents shall be available, upon request, to the department.

(2) Each subsidiary or branch laboratory shall independently comply with this section.

(3) At a minimum, the quality control program shall consist of:

(a) Calibration and maintenance of test instruments and equipment as necessary to maintain accuracy.

(b) A known standard analyzed or a calibration done on each analysis day. The instrument response for the known standard shall be within the pre-established limits under par. (c).

(c) A known standard analyzed after the analysis of 20 samples, if 20 or more samples are analyzed in an analysis day. The instrument response for the known standard shall be within the following pre-established limits:

1. For test categories 2, 3, 6, 9, 10 and for total organic carbon, total organic halide, chloride, hardness, and sulfate, the pre-established limit shall be ±10%.

2. For test categories 11, 12, 13, 14, 15, 16, 17, and 18, the pre-established limits shall be ±15%.

3. There is no requirement to analyze a known standard for alkalinity/acidity, corrosivity, EP toxicity, ignitability reactivity, gravimetric tests, titratimetric tests, and test categories 4 and 7.

4. For test category 1, a known standard shall be analyzed after the analysis of 20 samples. The limits on this quality control check shall be as established in an authoritative source.

5. For test category 20 the pre-established limit shall be appropriate for the test.

(d) At least one reagent blank shall be analyzed on each analysis day, for those tests for which reagent blanks are appropriate. For certain tests, a nonreacted sample may be used as a blank.

Note: Reagent blanks are not appropriate for certain tests such as alkalinity/acidity, conductivity, hardness, ignitability, and pH. Nonreacted sample blanks are appropriate for certain colormetric and turbidimetric tests such as the sulfate turbidimetric test, silica molybdosilicate test, and the phosphorus ascorbic acid test.

(e) A duplicate sample shall be run after the analysis of 10 samples. For those methodologies which require that the sample bottle be extracted, duplicate samples shall be taken in the field to insure representative samples.

(f) Spiked samples shall be analyzed except when the method of standard addition is used. The spiking of the sample shall be done before any extraction or digestion. The frequency of spiked analysis shall be:

1. As required in the authoritative sources for test categories 11 to 18, 20, total organic halide and total organic carbon. If no frequency is given in the authoritative source, then the frequency shall be after the analysis of 10 samples.

2. After the analysis of 20 samples, at a minimum, for test categories 2, 3, 6, 9, 10, and for chloride, hardness, sulfate, and bromide.

3. No spiked analysis is required for test categories 1 and 4 and for alkalinity/acidity, color, EP toxicity, pH, oil and grease, specific conductance, sulfide, sulfite, turbidity, corrosivity, ignitability, reactivity, and gravimetric tests, or tests where appropriate standards are not available for spiking.

Note: Spiked samples for organics should be spiked with representative organic analytes for each analyzed extraction. The representative organic analytes should be chosen on the basis of the organic analytes which are identified within a permit, and organic analytes which are typically found in that type of sample.

(g) Quality control limits for duplicate sample and spiked sample analysis shall be calculated using a method from an authoritative source. For laboratories with less than 30 quality control results within 12 months, the laboratory may set quality control limits based on information given in the authoritative sources, or laboratory experience, or the experience of other laboratories.

(h) If the results of known standards or duplicates exceed quality control limits, corrective action shall be taken by the laboratory. If it is determined by the laboratory that the discrepancy has affected past sample results, the laboratory shall reanalyze the samples or qualify the results back to the last acceptable quality control check. The results are qualified by reporting that the laboratory analysis was not within the acceptance limits for this test.

(i) If the analysis of a spiked sample exceeds the quality control limits, corrective action shall be taken by the laboratory. If it is determined by the laboratory that the discrepancy has affected past sample results, the laboratory shall reanalyze the samples or qualify the results, for those samples of the same sample matrix, back to the last acceptable quality control check. The results are qualified by reporting that the laboratory analysis was not within acceptance limits for this test. The impact of the spiked sample results on samples of different sample matrices shall be examined to insure that whatever affected the spiked sample had no impact on those samples of different matrices.

(j) A blind standard shall be analyzed, if available for that analyte, every 4 months for each analyte in test categories 1 to 10, 16, and 20, if the analyte was analyzed during the previous 4-month period. A blind standard shall be analyzed every 4 months for one analyte in each test category in test categories 11 to 15, 17, and 18, if an analyte within those test categories was analyzed during the previous 4-month period. If the result for any analyte does not fall within the limits established by the provider or the laboratory, corrective action shall be taken by the laboratory and an additional blind standard shall be analyzed to verify that the corrective action was successful.

(k) Where duplicate, spikes, and other quality control limits are exceeded, documentation shall be available to the department, upon request, indicating what corrective action was taken to bring the results back within limits.

(4) A copy of the methodology used by the laboratory for each analyte analyzed shall be available to the analyst.

TABLE 1
Test Categories

No.	Test Category	Key Analyte	Analytes In Test Category (Includes all forms of the given analytes)
1.	Oxygen Utilization	Total BOD5	Biochemical oxygen demand, carbonaceous biochemical oxygen demand
2.	Nitrogen	Each analyte for which certification or registration is desired except nitrite.	Nitrate as Nitrogen, Nitrite as Nitrogen, Ammonia as Nitrogen, total Kjeldahl Nitrogen.
3.	Phosphorus	Total Phosphorus	Orthophosphate, Phosphorus
4.	Physical	Total Suspended Solids	Total Solids, Dissolved Solids, Volatile Solids, Total Suspended Solids.
5.	General I	Chloride	Alkalinity/Acidity, Chloride, Hardness, Sulfate
6.	General II	Each analyte for which certification or registration is desired.	Chemical Oxygen Demand, Cyanide, Fluoride, Total Phenolic Compounds
7.	General III	No reference sample	Bromide, Color, odor, Oil and Grease, Specific Conductance, Sulfide, Sulfite, Surfactants, Turbidity
8.	General IV	No reference sample	Corrosivity, EP Toxicity, Ignitability, Reactivity, Total Organic Carbon, Total Organic Halide.
9.	Metals I	Copper and Cadmium	Aluminum, Antimony, Barium, Beryllium, Bismuth, Boron, Cadmium, Calcium, Chromium, Cobalt, Copper, Iron, Gold, Iridium, Lead, Lithium, Magnesium, Manganese, Molybdenum, Nickel, Osmium, Palladium, Platinum, Potassium, Rhodium, Ruthenium, Silicon, Silver, Sodium, Strontium, Thallium, Tin, Titanium, Tungsten, Vanadium, Zinc, and Zirconium.
10.	Metals II	Each analyte for which certification or registration is desired	Aluminum, Antimony, Arsenic, Barium, Beryllium, Bismuth, Boron, Cadmium, Calcium, Chromium, Cobalt, Copper, Iron, Gold, Iridium, Lead, Lithium, Magnesium, Manganese, Mercury, Molybdenum, Nickel, Osmium, Palladium, Platinum, Potassium, Rhodium, Ruthenium, Selenium, Silicon, Silver, Sodium, Strontium, Thallium, Tin, Titanium, Tungsten, Vanadium, Zinc, and Zirconium.
11.	Organics; Purgeable by Gas Chromatography or Gas Chromatography/Mass Spectrometer	Trichloroethene and Benzene	Purgeable Halocarbons, Purgeable Aromatics, Arolein, Acrylonitrile.

A T T A C H M E N T I I I

OPERATIONAL DATA SHEETS

DATE: / / .
PROJECT NO.: 291-012-29-39

NEW BEDFORD HARBOR - DATA LOG

Solids Analysis

REACTOR NUMBER	TIN NUMBER	TARE gms.	TARE + SAMPLE gms.	DRY gms.	ASHED gms.	TS %	VTS %

NOTES:

TS=Total Soilds
VTS=Volitile Total Solids

ANALYST

DATE: ___/___/___.
PROJECT NO.: 291-012-29-39

NEW BEDFORD HARBOR - DATA LOG

Oxygen Uptake Rates

REACTOR NUMBER	TIME	UPTAKE mg/L/hr	TIME	UPTAKE mg/L/hr	TIME	UPTAKE mg/L/hr	TIME	UPTAKE mg/L/hr

NOTES:

ANALYST

DATE: / / .
PROJECT NO.: 291-012-29-39

NEW BEDFORD HARBOR - DATA LOG

Reactor Weights

REACTOR NUMBER	INITIAL Wt., gms	VOLUME ml	FINAL Wt., gms	TARGET Wt., gms	TURBIDTY NTU	COLOR	COMMENTS

NOTES:

ANALYST

E X H I B I T A

SAMPLE SHIPPING LABEL

CHAIN-OF-CUSTODY SHEET

RADIAN CORPORATION

PROJECT NO. _____

RADIAN - 291-012-29-39
EBASCO - 4236-LAB-0062

SAMPLE I.D. -

PRESERVATIVE -

DESCRIPTION -

TYPE - ☐ Grab ☐ Composite

ENVIRONMENTAL PROTECTION AGENCY
Region I Waste Management Division

CHAIN OF CUSTODY RECORD

PROJ. NO.	PROJECT NAME	NO. OF CONTAINERS							REMARKS
SAMPLERS: *(Signature)*									
STA. NO.	DATE	TIME	COMP.	GRAB	STATION LOCATION				

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Relinquished by: *(Signature)*	Date / Time	Received by: *(Signature)*	Relinquished by: *(Signature)*	Date / Time	Received by: *(Signature)*
Relinquished by: *(Signature)*	Date / Time	Received by: *(Signature)*	Relinquished by: *(Signature)*	Date / Time	Received by: *(Signature)*
Relinquished by: *(Signature)*	Date / Time	Received for Laboratory by: *(Signature)*	Date / Time	Remarks	

Distribution: Original Accompanies Shipment; Copy to Coordinator Field Files

02996

APPENDIX D

DATA FOR CULTURE GROWTH PARAMETERS

TABLE D-1. SAMPLE IDENTIFICATION KEY

REACTOR NUMBER (FROM TABLE D-2)	DESCRIPTION
1-6	Biphenyl culture development reactors.
7	Replacement for flask No. 1 which broke.
1A-6A	Biphenyl first growth confirmation reactors.
Blank (Days 18-30)	During the growth confirmation phase, this reactor contained media (seawater and nutrietns) and biphenyl, but no seed.
Blank (Days 82-96)	Formaldehyde analysis; blank contained dionized water and reagents.
Sediment HL	High PCB level harbor sediment - as received.
Sediment LL	Low PCB level harbor sediment - as received.
1B-6B	Biphenyl second growth confirmation reactors.
7R-8R	Biphenyl first growth reactors setup to confirm results of reactors 1A-6A.
7RA-8RA	Biphenyl second growth confirmation.
7A	Combined contents of reactors 2 and 3 (biphenyl culture development phase).
8A	Combined contents of reactors 4 and 6.
HL Media	High PCB level media used to feed PCB culture reactors.
LL Media	Low PCB level media used to feed PCB culture reactors.
1H-4H	High level media reactors - PCB culture development phase.
5L-7L	Low level media reactors - PCB culture development phase.

TABLE D-1 (Continued)

5L'-6L'	Low level media reactors. These reactors replaced 5L and 6L on May 3, 1988.
1H-4H (Composite)	Presumptive phase, 10-day effluent composite.
Influent Comp.	Presumptive phase, 10-day influent composite.
5.41 STD	Prepared standard containing 5.41 mg/l of formaldehyde.
7CCH	High level confirmation phase control reactor (formaldehyde added).
7CCHSPK	Contents of reactor 7CCH to which a known amunt of standard solution was added.
C1H-C4H	High PCB level confirmation phase reactors.
C5L-C6L	Low PCB level confirmation phase reactors.
8CCL	Low PCB level confirmation phase control reactor (formaldehyde added).

TABLE D-2
NEW BEDFORD PCB PROJECT
DATA SUMMARY-RADIAN

Day	Date	Reactor Number	D.O. mg/L	O.U. mg/L/hr	T.S. ppm	T.V.S. ppm	TURBIDITY NTU	HANTZSCH FORMALDEHYDE %
		BIPHENYL CULTURE DEVELOPMENT						
1	02/22/88	1&2	7.2	3.53				
1	02/22/88	3&4	6.7	3.39				
1	02/22/88	5&6	6.6	3.29				
3	02/24/88	2&3	6.4	22.8				
3	02/24/88	4&5	6.2	26.4				
3	02/24/88	1&6	5.69	21.6				
2	02/23/88	1			4530	634		
2	02/23/88	1			4830	687		
2	02/23/88	2			4770	641		
2	02/23/88	2			4740	692		
2	02/23/88	3			4790	659		
2	02/23/88	3			4560	593		
2	02/23/88	4			4960	756		
2	02/23/88	4			4880	708		
2	02/23/88	5			4780	643		
2	02/23/88	5			4720	691		
2	02/23/88	6			4950	636		
2	02/23/88	6			4860	648		
4	02/25/88	1			4320	919		
4	02/25/88	1			4310	935		
4	02/25/88	2			4530	910		
4	02/25/88	2			4580	922		
4	02/25/88	3			4480	903		
4	02/25/88	3			4490	894		
4	02/25/88	4			4780	1020		
4	02/25/88	4			4860	1040		
4	02/25/88	5			4500	980		
4	02/25/88	5			4440	993		
4	02/25/88	6			4360	941		
4	02/25/88	6			4320	922		
8	02/29/88	3&4	7.2	40				
8	02/29/88	2&5	5	40				
8	02/29/88	1&6	5.85	23				
9	03/01/88	1			5370	1490		
9	03/01/88	1			5270	1480		
9	03/01/88	2			6570	1680		
9	03/01/88	2			6330	1690		
9	03/01/88	3			6820	1840		
9	03/01/88	3			6870	1870		

TABLE D-2
NEW BEDFORD PCB PROJECT
DATA SUMMARY-RADIAN

Day	Date	Reactor Number	D.O. mg/L	O.U. mg/L/hr	T.S. ppm	T.V.S. ppm	TURBIDITY NTU	HANTZSCH FORMALDEHYDE %
9	03/01/88	4			6650	1770		
9	03/01/88	4			6590	1790		
9	03/01/88	5			6030	1610		
9	03/01/88	5			6070	1620		
9	03/01/88	6			6290	1650		
9	03/01/88	6			6140	1630		
11	03/03/88	1			8710	1570		
11	03/03/88	1			8720	1590		
11	03/03/88	2			9110	1530		
11	03/03/88	2			8960	1520		
11	03/03/88	3			8700	1490		
11	03/03/88	3			8600	1510		
11	03/03/88	4			9160	1610		
11	03/03/88	4			9310	1610		
11	03/03/88	5			8900	1490		
11	03/03/88	5			8780	1530		
11	03/03/88	6			9480	1650		
11	03/03/88	6			9350	1680		
10	03/02/88	1&3	7.2	25.7				
10	03/02/88	2&5	6.4	32				
10	03/02/88	4&6	5.7	32				
15	03/07/88	1&2	5.6	35				
15	03/07/88	3&6	4	37				
15	03/07/88	4&5	5.6	39				
16	03/08/88	1			12400	1610		
16	03/08/88	1			12400	1600		
16	03/08/88	2			12600	1650		
16	03/08/88	2			12600	1650		
16	03/08/88	3			13400	1820		
16	03/08/88	3			13300	1690		
16	03/08/88	4			13500	1790		
16	03/08/88	4			13500	1760		
16	03/08/88	5			13000	1690		
16	03/08/88	5			13100	2050		
16	03/08/88	6			13200	1840		
16	03/08/88	6			13100	1750		
17	03/09/88	1	7.2					
17	03/09/88	2	7					
17	03/09/88	5	7.2					
17	03/09/88	2&3	6	30				
17	03/09/88	7&5	6.2	26				
17	03/09/88	4&6	6	30				

TABLE D-2
NEW BEDFORD PCB PROJECT
DATA SUMMARY-RADIAN

Day	Date	Reactor Number	D.O. mg/L	O.U. mg/L/hr	T.S. ppm	T.V.S. ppm	TURBIDITY NTU	HANTZSCH FORMALDEHYDE %
		BIPHENYL FIRST GROWTH CONFIRMATION						
17	03/09/88	1A					1.4	
17	03/09/88	2A					1.1	
17	03/09/88	3A					2.4	
17	03/09/88	4A					0.75	
17	03/09/88	5A					0.8	
17	03/09/88	6A					0.5	
18	03/10/88	1A					3.4	
18	03/10/88	2A					2.3	
18	03/10/88	3A					4.8	
18	03/10/88	4A					2.6	
18	03/10/88	5A					2.7	
18	03/10/88	6A					2.6	
18	03/10/88	BLANK					0.75	
18	03/10/88	BLANK					0.75	
19	03/11/88	1A					5.4	
19	03/11/88	2A					4.5	
19	03/11/88	3A					7	
19	03/11/88	4A					5.2	
19	03/11/88	5A					5.3	
19	03/11/88	6A					4.4	
19	03/11/88	BLANK					0.75	
20	03/12/88	1A					6.2	
20	03/12/88	2A					6.3	
20	03/12/88	3A					10.2	
20	03/12/88	4A					8.2	
20	03/12/88	5A					8.4	
20	03/12/88	6A					7.8	
20	03/12/88	BLANK					0.75	
22	03/14/88	SEDIMENT HL			301000			
22	03/14/88	SEDIMENT LL			369000			
22	03/14/88	1A					3.2	
22	03/14/88	2A					5.5	
22	03/14/88	3A					15	
22	03/14/88	4A					14	
22	03/14/88	5A					14	
22	03/14/88	6A					32	
22	03/14/88	BLANK					3.5	
22	03/14/88	7A	6.2	35				
22	03/14/88	8A	3.6	38				

TABLE D-2
NEW BEDFORD PCB PROJECT
DATA SUMMARY-RADIAN

Day	Date	Reactor Number	D.O. mg/L	O.U. mg/L/hr	T.S. ppm	T.V.S. ppm	TURBIDITY NTU	HANTZSCH FORMALDEHYDE %
23	03/15/88	1A					8.2	
23	03/15/88	2A					11	
23	03/15/88	3A					24	
23	03/15/88	4A					21	
23	03/15/88	5A					21	
23	03/15/88	6A					23	
23	03/15/88	BLANK					5 *	
		BIPHENYL SECOND GROWTH CONFIRMATION						
23	03/15/88	1B					0.42	
23	03/15/88	2B					0.33	
23	03/15/88	3B					0.57	
23	03/15/88	4B					0.43	
23	03/15/88	5B					0.54	
23	03/15/88	6B					0.97	
23	03/15/88	7R					2.8	
23	03/15/88	8R					3.7	
23	03/15/88	BLANK					0.74 **	
23	03/15/88	7A			14700	2050		
23	03/15/88	7A			14600	2000		
23	03/15/88	8A			15200	2270		
23	03/15/88	8A			15100	2270		
24	03/16/88	1B					0.62	
24	03/16/88	2B					0.98	
24	03/16/88	3B					1.2	
24	03/16/88	4B					1.3	
24	03/16/88	5B					1.2	
24	03/16/88	6B					2	
24	03/16/88	BLANK					4.75	
24	03/16/88	1A					12	
24	03/16/88	3A					28	
24	03/16/88	7R					5.4	
24	03/16/88	8R					6.9	
24	03/16/88	BLANK					0.68	
25	03/17/88	1B					3	
25	03/17/88	2B					1.5	
25	03/17/88	3B					2.5	
25	03/17/88	4B					1.5	
25	03/17/88	5B					1.75	
25	03/17/88	6B					4.2	
25	03/17/88	BLANK					6 *	

*This blank discarded
**New blank prepared

TABLE D-2
NEW BEDFORD PCB PROJECT
DATA SUMMARY-RADIAN

Day	Date	Reactor Number	D.O. mg/L	O.U. mg/L/hr	T.S. ppm	T.V.S. ppm	TURBIDITY NTU	HANTZSCH FORMALDEHYDE %
25	03/17/88	1A					16	
25	03/17/88	3A					32	
25	03/17/88	7R					15	
25	03/17/88	8R					17	
25	03/17/88	BLANK					0.7 **	
26	03/18/88	1B					3.5	
26	03/18/88	2B					2.2	
26	03/18/88	3B					2.5	
26	03/18/88	4B					3.2	
26	03/18/88	5B					2.4	
26	03/18/88	6B					6.1	
26	03/18/88	BLANK					5.5 **	
26	03/18/88	1A					18	
26	03/18/88	3A					36	
26	03/18/88	7R					21	
26	03/18/88	8R					23	
26	03/18/88	7RA					1.4	
26	03/18/88	8RA					1.3	
26	03/18/88	BLANK					0.81 *	
27	03/19/88	1B					3.2	
27	03/19/88	2B					2.5	
27	03/19/88	3B					2.7	
27	03/19/88	4B					3.1	
27	03/19/88	5B					1.3	
27	03/19/88	6B					6.3	
27	03/19/88	BLANK					2.4	
27	03/19/88	1A					20	
27	03/19/88	3A					40	
27	03/19/88	7RA					4.2	
27	03/19/88	8RA					2.4	
29	03/21/88	1B					4.5	
29	03/21/88	2B					4.8	
29	03/21/88	3B					14	
29	03/21/88	4B					10	
29	03/21/88	5B					4.4	
29	03/21/88	6B					23	
29	03/21/88	BLANK					8.3	
29	03/21/88	1A					26	
29	03/21/88	3A					45	
29	03/21/88	7RA					25	
29	03/21/88	8RA					27	
30	03/22/88	HL MEDIA			21100	2690		

*This blank discarded
**New blank prepared

TABLE D-2
NEW BEDFORD PCB PROJECT
DATA SUMMARY-RADIAN

Day	Date	Reactor Number	D.O. mg/L	O.U. mg/L/hr	T.S. ppm	T.V.S. ppm	TURBIDITY NTU	HANTZSCH FORMALDEHYDE %
30	03/22/88	HL MEDIA			20900	2530		
30	03/22/88	LL MEDIA			22200	2110		
30	03/22/88	LL MEDIA			22100	1980		
30	03/22/88	1B					6.2	
30	03/22/88	2B					5.8	
30	03/22/88	3B					13	
30	03/22/88	4B					10	
30	03/22/88	5B					6	
30	03/22/88	6B					25	
30	03/22/88	BLANK					13	
30	03/22/88	1A					28	
30	03/22/88	3A					48	
30	03/22/88	7RA					25	
30	03/22/88	8RA					29	
30	03/22/88	7A		47				
30	03/22/88	8A		49				
		PCB CULTURE DEVELOPMENT						
31	03/23/88	1H	7.6					
31	03/23/88	2H	7.1	1.3				
31	03/23/88	3H	7.1					
31	03/23/88	4H	6.8	0.9				
31	03/23/88	5L	7.4					
31	03/23/88	6L	7.4	0.9				
31	03/23/88	7L	8					
31	03/23/88	7A			14900	2530		
31	03/23/88	7A			14800	2460		
31	03/23/88	8A			14800	2780		
31	03/23/88	8A			15000	2800		
32	03/24/88	1H			19700	2290		
32	03/24/88	1H			19700	2130		
32	03/24/88	2H			19700	2200		
32	03/24/88	2H			19900	2910		
32	03/24/88	3H			18800	2190		
32	03/24/88	3H			18700	1980		
32	03/24/88	4H			19400	2150		
32	03/24/88	4H			19500	2050		
32	03/24/88	5L			21400	2050		
32	03/24/88	5L			21400	2090		
32	03/24/88	6L			21000	1910		
32	03/24/88	6L			20800	2040		

TABLE D-2
NEW BEDFORD PCB PROJECT
DATA SUMMARY-RADIAN

Day	Date	Reactor Number	D.O. mg/L	O.U. mg/L/hr	T.S. ppm	T.V.S. ppm	TURBIDITY NTU	HANTZSCH FORMALDEHYDE %
32	03/24/88	7L			21200	2040		
32	03/24/88	7L			21300	2110		
36	03/28/88	1H	6.4					
36	03/28/88	2H	6					
36	03/28/88	3H	6.3					
36	03/28/88	4H	6.4					
36	03/28/88	5L	5.5					
36	03/28/88	6L	4.1					
36	03/28/88	7L	4.3					
36	03/28/88	7A		38.6				
36	03/28/88	8A		31.6				
37	03/29/88	1H			19400	2870		
37	03/29/88	1H			18900	2700		
37	03/29/88	2H			21000	3380		
37	03/29/88	2H			20400	3180		
37	03/29/88	3H			20400	3350		
37	03/29/88	3H			20700	3110		
37	03/29/88	4H			19600	3005		
37	03/29/88	4H			19500	2920		
37	03/29/88	5L			20300	2770		
37	03/29/88	5L			21000	3020		
37	03/29/88	6L			20600	2960		
37	03/29/88	6L			22000	3270		
37	03/29/88	7L			21600	3200		
37	03/29/88	7L			20800	3000		
37	03/29/88	7A			13000	1870		
37	03/29/88	7A			13000	1910		
37	03/29/88	8A			13300	2060		
37	03/29/88	8A			13100	2050		
38	03/30/88	1H	8.4	11.7				
38	03/30/88	2H	7.2	10.2				
38	03/30/88	3H	8	8.9				
38	03/30/88	4H	7.1	7.2				
38	03/30/88	5L	7.2	8.4				
38	03/30/88	6L	7.7	8.24				
38	03/30/88	7L	7	10.8				
39	03/31/88	1H			18500	2700		
39	03/31/88	1H			19100	2830		
39	03/31/88	2H			19100	2830		
39	03/31/88	2H			19400	2970		
39	03/31/88	3H			18700	2870		
39	03/31/88	3H			19900	3170		

TABLE D-2
NEW BEDFORD PCB PROJECT
DATA SUMMARY-RADIAN

Day	Date	Reactor Number	D.O. mg/L	O.U. mg/L/hr	T.S. ppm	T.V.S. ppm	TURBIDITY NTU	HANTZSCH FORMALDEHYDE %
39	03/31/88	4H			19600	3030		
39	03/31/88	4H			21400	3250		
39	03/31/88	5L			20900	2940		
39	03/31/88	5L			21900	3230		
39	03/31/88	6L			20700	2780		
39	03/31/88	6L			21000	2900		
39	03/31/88	7L			20500	2730		
39	03/31/88	7L			20700	2750		
39	03/31/88	HL MEDIA			19500	2460		
39	03/31/88	HL MEDIA			19800	2420		
43	04/04/88	1H	8.4					
43	04/04/88	2H	7.6					
43	04/04/88	3H	8.4					
43	04/04/88	4H	9.1					
43	04/04/88	5L	8					
43	04/04/88	6L	11.7					
43	04/04/88	7L	9.5					
43	04/04/88	7A		24.5				
43	04/04/88	8A		31.6				
44	04/05/88	1H			18700	2720		
44	04/05/88	1H			19200	2830		
44	04/05/88	2H			19500	2960		
44	04/05/88	2H			19000	2710		
44	04/05/88	3H			19600	3010		
44	04/05/88	3H			19400	2880		
44	04/05/88	4H			21100	3410		
44	04/05/88	4H			20400	3090		
44	04/05/88	5L			19700	2600		
44	04/05/88	5L			20300	2790		
44	04/05/88	6L			20666	2750		
44	04/05/88	6L			20200	2620		
44	04/05/88	7L			20500	2810		
44	04/05/88	7L			20800	2740		
44	04/05/88	7A			14000	1920		
44	04/05/88	7A			14100	1890		
44	04/05/88	8A			14000	2000		
44	04/05/88	8A			13900	1960		
45	04/06/88	1H	7.8	6.7				
45	04/06/88	2H	7.5	4.74				
45	04/06/88	3H	6.9	4.62				
45	04/06/88	4H	8	2.31				
45	04/06/88	5L	6.8	4.74				

TABLE D-2
NEW BEDFORD PCB PROJECT
DATA SUMMARY-RADIAN

Day	Date	Reactor Number	D.O. mg/L	O.U. mg/L/hr	T.S. ppm	T.V.S. ppm	TURBIDITY NTU	HANTZSCH FORMALDEHYDE %
45	04/06/88	6L	7.1	4.62				
45	04/06/88	7L	7.2	5				
46	04/07/88	1H			17200	2330		
46	04/07/88	1H			17500	2500		
46	04/07/88	2H			20800	3140		
46	04/07/88	2H			19700	2930		
46	04/07/88	3H			19500	3020		
46	04/07/88	3H			19100	2880		
46	04/07/88	4H			20100	2880		
46	04/07/88	4H			19800	2770		
46	04/07/88	5L			20500	2760		
46	04/07/88	5L			21900	2890		
46	04/07/88	6L			24600	3320		
46	04/07/88	6L			22600	4251		
46	04/07/88	7L			18300	2460		
46	04/07/88	7L			19200	1440		
47	04/08/88	1H		4.2 @ 7:05AM				
47	04/08/88	1H		6.8 @ 11:00AM				
47	04/08/88	1H		7.06 @ 1:00PM				
47	04/08/88	1H		6.2 @ 3:40PM				
47	04/08/88	7L		7.27 @ 7:05AM				
47	04/08/88	7L		7.5 @ 11:00AM				
47	04/08/88	7L		7.06 @ 1:00PM				
47	04/08/88	7L		6.9 @ 3:40PM				
50	04/11/88	1H	7.6					
50	04/11/88	2H	7.7					
50	04/11/88	3H	7.6					
50	04/11/88	4H	7.6					
50	04/11/88	5L	7.7					
50	04/11/88	6L	7.05					
50	04/11/88	7L	6.6					
50	04/11/88	7A		33.8				
50	04/11/88	8A		26.9				
51	04/12/88	1H			16600	2390		
51	04/12/88	1H			17200	2540		
51	04/12/88	2H			20600	3220		
51	04/12/88	2H			21400	3620		
51	04/12/88	3H			20000	3000		
51	04/12/88	3H			20600	3210		
51	04/12/88	4H			20300	3030		
51	04/12/88	4H			20900	2850		
51	04/12/88	5L			20400	2790		

TABLE D-2
NEW BEDFORD PCB PROJECT
DATA SUMMARY-RADIAN

Day	Date	Reactor Number	D.O. mg/L	O.U. mg/L/hr	T.S. ppm	T.V.S. ppm	TURBIDITY NTU	HANTZSCH FORMALDEHYDE %
51	04/12/88	5L			20000	2530		
51	04/12/88	6L			20800	2890		
51	04/12/88	6L			21100	3080		
51	04/12/88	7A			15100	2190		
51	04/12/88	7A			15200	2240		
51	04/12/88	8A			15600	2210		
51	04/12/88	8A			14700	2410		
51	04/12/88	LL MEDIA			21400	2740		
51	04/12/88	LL MEDIA			21400	2790		
52	04/13/88	1H	8.6	8.33				
52	04/13/88	2H	8.65	8.11				
52	04/13/88	3H	9.05	10.9				
52	04/13/88	4H	9.1	9.23				
52	04/13/88	5L	8.9	8.9				
52	04/13/88	6L	8.23	8.58				
52	04/13/88	7A		38.2				
52	04/13/88	8A		34.3				
53	04/14/88	1H			17500	2550		
53	04/14/88	1H			16600	2480		
53	04/14/88	2H			20100	3060		
53	04/14/88	2H			20400	3240		
53	04/14/88	3H			19100	1550		
53	04/14/88	3H			17900	1750		
53	04/14/88	4H			20700	4680		
53	04/14/88	4H			22000	4800		
53	04/14/88	5L			19900	2590		
53	04/14/88	5L			20100	2830		
53	04/14/88	6L			20600	2820		
53	04/14/88	6L			21100	2960		
57	04/18/88	7A		33.6				
57	04/18/88	8A		40.8				
58	04/19/88	1H			18300	2500		
58	04/19/88	1H			17800	2450		
58	04/19/88	2H			21500	3261		
58	04/19/88	2H			20900	3092		
58	04/19/88	3H			19600	2860		
58	04/19/88	3H			20900	3110		
58	04/19/88	4H			21200	2880		
58	04/19/88	4H			21100	3010		
58	04/19/88	5L			21000	2870		
58	04/19/88	5L			20800	2780		
58	04/19/88	6L			20600	2800		

TABLE D-2
NEW BEDFORD PCB PROJECT
DATA SUMMARY-RADIAN

Day	Date	Reactor Number	D.O. mg/L	O.U. mg/L/hr	T.S. ppm	T.V.S. ppm	TURBIDITY NTU	HANTZSCH FORMALDEHYDE %
58	04/19/88	6L			20900	2790		
58	04/19/88	7A			15900	3310		
58	04/19/88	7A			17100	3170		
58	04/19/88	8A			15200	2610		
58	04/19/88	8A			14100	2620		
59	04/20/88	1H	7.6	6.43				
59	04/20/88	2H	7	6.43				
59	04/20/88	3H	7.3	8.28				
59	04/20/88	4H	7.3	8.86				
59	04/20/88	5L	6.3	9				
59	04/20/88	6L	7	8.57				
60	04/21/88	1H			18100	2480		
60	04/21/88	1H			18700	2650		
60	04/21/88	2H			20500	3080		
60	04/21/88	2H			20900	3080		
60	04/21/88	3H			20200	3150		
60	04/21/88	3H			20700	3090		
60	04/21/88	4H			20700	2970		
60	04/21/88	4H			21800	3400		
60	04/21/88	5L			20700	2960		
60	04/21/88	5L			20400	2920		
60	04/21/88	6L			20300	2770		
60	04/21/88	6L			20200	2840		
64	04/25/88	1H	8.7					
64	04/25/88	2H	9.05					
64	04/25/88	3H	8.85					
64	04/25/88	4H	9.1					
64	04/25/88	5L	8.85					
64	04/25/88	6L	9.05					
64	04/25/88	7A		62.3				
64	04/25/88	8A		53				
65	04/26/88	1H			18900	2700		
65	04/26/88	1H			19400	2890		
65	04/26/88	2H			21300	3250		
65	04/26/88	2H			21000	3050		
65	04/26/88	3H			21600	3370		
65	04/26/88	3H			21400	3190		
65	04/26/88	4H			22800	3640		
65	04/26/88	4H			22200	3390		
65	04/26/88	5L			20900	2900		
65	04/26/88	5L			20600	2940		
65	04/26/88	6L			21100	3010		

TABLE D-2
NEW BEDFORD PCB PROJECT
DATA SUMMARY-RADIAN

Day	Date	Reactor Number	D.O. mg/L	O.U. mg/L/hr	T.S. ppm	T.V.S. ppm	TURBIDITY NTU	HANTZSCH FORMALDEHYDE %
65	04/26/88	6L			20600	2900		
65	04/26/88	7A			16600	2910		
65	04/26/88	7A			16700	2980		
65	04/26/88	8A			16500	3730		
65	04/26/88	8A			16600	3990		
66	04/27/88	1H	8.6	9.2				
66	04/27/88	2H	8.6	7.5				
66	04/27/88	3H	8.6	9.1				
66	04/27/88	4H	8.4	7.3				
66	04/27/88	5L	8.3	8.6				
66	04/27/88	6L	8.5	10.2				
67	04/28/88	1H			19800	2950		
67	04/28/88	1H			19500	2800		
67	04/28/88	2H			21500	3200		
67	04/28/88	2H			22100	3300		
67	04/28/88	3H			21600	3280		
67	04/28/88	3H			22200	3420		
67	04/28/88	4H			22700	3500		
67	04/28/88	4H			22500	3380		
67	04/28/88	5L			21200	3120		
67	04/28/88	5L			21300	3100		
67	04/28/88	6L			21400	3050		
67	04/28/88	6L			21500	2970		
		HIGH LEVEL PRESUMPTIVE TEST						
71	05/02/88	7A		34.3				
71	05/02/88	8A		38.2				
		LOW LEVEL REACCLIMATION						
72	05/03/88	7A			18200	3520		
72	05/03/88	7A			18200	3530		
72	05/03/88	8A			19200	4750		
72	05/03/88	8A			19100	4660		
72	05/03/88	LL MEDIA			23100	2820		
72	05/03/88	LL MEDIA			23700	2960		
72	05/03/88	HL MEDIA			22900	3210		
72	05/03/88	HL MEDIA			22500	3240		
73	05/04/88	5L'	10.4					
73	05/04/88	6L'	9.9					
74	05/05/88	5L'			23700	3140		

TABLE D-2
NEW BEDFORD PCB PROJECT
DATA SUMMARY-RADIAN

Day	Date	Reactor Number	D.O. mg/L	O.U. mg/L/hr	T.S. ppm	T.V.S. ppm	TURBIDITY NTU	HANTZSCH FORMALDEHYDE %
74	05/05/88	5L'			27800	3540		
74	05/05/88	6L'			22400	2920		
74	05/05/88	6L'			23100	3040		
75	05/06/88	5L'	7.5	12.4				
75	05/06/88	6L'	6.6	15				
76	05/07/88	5L'			23400	3320		
76	05/07/88	5L'			23100	3200		
76	05/07/88	6L'			22300	3150		
76	05/07/88	6L'			22700	3310		
78	05/09/88	5L'	7.9					
78	05/09/88	6L'	8.1					
78	05/09/88	7A		4 1:40PM				
78	05/09/88	8A		43.3 1:55PM				
78	05/09/88	7A		11.1 2:05PM				
79	05/10/88	5L'			24900	3670		
79	05/10/88	5L'			24100	3440		
79	05/10/88	6L'			26400	3680		
79	05/10/88	6L'			29400	3800		
79	05/10/88	7A			19600	3980		
79	05/10/88	7A			19600	4050		
79	05/10/88	8A			20200	4930		
79	05/10/88	8A			20300	4900		
79	05/10/88	1H		10				
79	05/10/88	2H		13.7				
79	05/10/88	3H		12.2				
79	05/10/88	4H		12.9				
79	05/10/88	7A		11.9				
81	05/12/88	5L'			23200	3270		
81	05/12/88	5L'			23900	3220		
81	05/12/88	6L'			23800	3420		
81	05/12/88	6L'			23800	3370		
81	05/12/88	1H(Composite)			21500	3090		
81	05/12/88	1H(Composite)			21100	3100		
81	05/12/88	2H(Composite)			22300	3510		
81	05/12/88	2H(Composite)			21900	3410		
81	05/12/88	3H(Composite)			22100	3460		
81	05/12/88	3H(Composite)			23100	3770		
81	05/12/88	4H(Composite)			22500	3410		
81	05/12/88	4H(Composite)			23100	3710		
81	05/12/88	Influent Composite			22600	2990		
81	05/12/88	Influent Composite			23900	3300		

TABLE D-2
NEW BEDFORD PCB PROJECT
DATA SUMMARY-RADIAN

Day	Date	Reactor Number	D.O. mg/L	O.U. mg/L/hr	T.S. ppm	T.V.S. ppm	TURBIDITY NTU	HANTZSCH FORMALDEHYDE %
		HIGH LEVEL CONFIRMATION PHASE						
82	05/13/88	5L'	8.35	11.3				
82	05/13/88	6L'	8.1	7.2				
82	05/13/88	BLANK						0.11(mg/L)
82	05/13/88	5.41STD						5.35(mg/L)
82	05/13/88	7CCH						1.14
82	05/13/88	7CCH						1.24
82	05/13/88	7CCHSPK						1.85
83	05/14/88	5L'			22300	3150		
83	05/14/88	5L'			23900	3440		
83	05/14/88	6L'			23300	3530		
83	05/14/88	6L'			23100	3440		
83	05/14/88	BLANK						0.10(mg/L)
83	05/14/88	5.41STD						4.84(mg/L)
83	05/14/88	7CCH						1.3
83	05/14/88	7CCH						1.44
83	05/14/88	7CCHSPK						1.88
85	05/16/88	5L'	8.9					
85	05/16/88	6L'	9.05					
85	05/16/88	1H	10.2					
85	05/16/88	2H	10.2					
85	05/16/88	3H	10.4					
85	05/16/88	4H	9.85					
85	05/16/88	7A		4.1				
85	05/16/88	8A		5				
85	05/16/88	BLANK						0.18(mg/L)
85	05/16/88	5.41STD						4.82(mg/L)
85	05/16/88	7CCH						2.77
85	05/16/88	7CCH						2.47
85	05/16/88	7CCHSPK						4.71
86	05/17/88	5L'			23500	3300		
86	05/17/88	5L'			24100	3460		
86	05/17/88	6L'			22800	3310		
86	05/17/88	6L'			23900	3470		
86	05/17/88	7A			16300	2530		
86	05/17/88	7A			16700	2690		
86	05/17/88	8A			17900	4020		
86	05/17/88	8A			18000	4110		
86	05/17/88	BLANK						0.12(mg/L)
86	05/17/88	5.41STD						5.29(mg/L)
86	05/17/88	7CCH						OUT OF RANGE*

*Out of range of calibration curve

TABLE D-2
NEW BEDFORD PCB PROJECT
DATA SUMMARY-RADIAN

Day	Date	Reactor Number	D.O. mg/L	O.U. mg/L/hr	T.S. ppm	T.V.S. ppm	TURBIDITY NTU	HANTZSCH FORMALDEHYDE %
86	05/17/88	7CCH						OUT OF RANGE*
86	05/17/88	7CCHSPK						OUT OF RANGE*
86	05/17/88	C1H			19500	3220		
86	05/17/88	C1H			23000	3120		
86	05/17/88	C2H			21600	3220		
86	05/17/88	C2H			21100	3190		
86	05/17/88	C3H			22200	3350		
86	05/17/88	C3H			21400	3140		
86	05/17/88	C4H			22100	3300		
86	05/17/88	C4H			23100	3550		
86	05/17/88	7CCH			22400	3260		
86	05/17/88	7CCH			22900	3180		
87	05/18/88	5L'	13.2	6.23				
87	05/18/88	6L'	13	5.14				
87	05/18/88	BLANK						0.10(mg/L)
87	05/18/88	5.41STD						4.86(mg/L)
87	05/18/88	7CCH						3.92
87	05/18/88	7CCH						3.44
87	05/18/88	7CCHSPK						4.42
87	05/18/88	C1H						0.04 *
88	05/19/88	5L'			22100	3090		
88	05/19/88	5L'			23400	3370		
88	05/19/88	6L'			22500	3410		
88	05/19/88	6L'			22200	3250		
88	05/19/88	BLANK						0.20(mg/L)
88	05/19/88	5.41STD						5.30(MG/L)
88	05/19/88	7CCH						1.52
88	05/19/88	7CCH						1.5
88	05/19/88	7CCHSPK						3.44
88	05/19/88	C2H						0.05 *
88	05/19/88	BLANK						0.06
88	05/19/88	5.41STD						5.24
88	05/19/88	7CCH						OUT OF RANGE
88	05/19/88	7CCH						2.44
88	05/19/88	7CCHSPK						3.84
88	05/19/88	C2H						0.04 *
89	05/20/88	LL MEDIA			17800	2650		
89	05/20/88	LL MEDIA			18200	2650		
89	05/20/88	BLANK						0.06(mg/L)
89	05/20/88	5.41STD						5.26(mg/L)
89	05/20/88	7CCH						2.08
89	05/20/88	7CCH						2.05

*"Active" reactors checked for formaldehyde contamination

TABLE D-2
NEW BEDFORD PCB PROJECT
DATA SUMMARY-RADIAN

Day	Date	Reactor Number	D.O. mg/L	O.U. mg/L/hr	T.S. ppm	T.V.S. ppm	TURBIDITY NTU	HANTZSCH FORMALDEHYDE %
89	05/20/88	7CCHSPK						3.58
89	05/20/88	C3H						0.12 *
92	05/23/88	5L'	8.92					
92	05/23/88	6L'	8.46					
92	05/23/88	7A		28.5				
92	05/23/88	8A		35.3				
92	05/23/88	7CCH						2.38
92	05/23/88	7CCH						2.38
92	05/23/88	7CCHSPK						2.57
93	05/24/88	5L'			20800	2800		
93	05/24/88	5L'			21200	2930		
93	05/24/88	6L'			20300	2830		
93	05/24/88	6L'			20800	3050		
93	05/24/88	7A			15800	2470		
93	05/24/88	7A			15800	2480		
93	05/24/88	8A			16100	3160		
93	05/24/88	8A			16200	3150		
93	05/24/88	BLANK						0.06(mg/L)
93	05/24/88	5.41STD						5.36(mg/L)
93	05/24/88	7CCH						2.12
93	05/24/88	7CCH						2.19
93	05/24/88	7CCHSPK						2.46
94	05/25/88	5L'	10.6	4.3				
94	05/25/88	6L'	10.3	1.5				
94	05/25/88	BLANK						0.06(mg/L)
94	05/25/88	5.41STD						5.20(mg/L)
94	05/25/88	7CCH						2.2
94	05/25/88	7CCH						2.22
94	05/25/88	7CCHSPK						2.4
95	05/26/88	5L'			19800	2620		
95	05/26/88	5L'			19500	2590		
95	05/26/88	6L'			20700	2950		
95	05/26/88	6L'			20200	2920		
95	05/26/88	BLANK						0.03(mg/L)
95	05/26/88	5.41STD						5.30(mg/L)
95	05/26/88	7CCH						2.17
95	05/26/88	7CCH						2.17
95	05/26/88	7CCHSPK						2.43
95	05/26/88	7CCH						
95	05/26/88	7CCH						
95	05/26/88	7CCH						
96	05/27/88	BLANK						-0.06(mg/L)

TABLE D-2
NEW BEDFORD PCB PROJECT
DATA SUMMARY-RADIAN

Day	Date	Reactor Number	D.O. mg/L	O.U. mg/L/hr	T.S. ppm	T.V.S. ppm	TURBIDITY NTU	HANTZSCH FORMALDEHYDE %
96	05/27/88	5.41STD						5.24(mg/L)
96	05/27/88	7CCH						2.37
96	05/27/88	7CCH						2.4
96	05/27/88	7CCHSPK						2.52
100	05/31/88	7CCH			20200	2940		
100	05/31/88	7CCH			20600	3030		
100	05/31/88	C2H			21900	3700		
100	05/31/88	C2H			21900	3800		
100	05/31/88	C3H			20900	3460		
100	05/31/88	C3H			21400	3490		
100	05/31/88	C4H			20500	3140		
100	05/31/88	C4H			19900	3100		
100	05/31/88	C1H			20600	3310		
100	05/31/88	C1H			20600	3110		
100	05/31/88	5L'	8.98					
100	05/31/88	6L'	9.5					
100	05/31/88	7A		19.15				
100	05/31/88	8A		25.12				
101	06/01/88	5L'		10.71	17900	2530		
101	06/01/88	5L'			17000	2390		
101	06/01/88	6L'		8.42	18300	2600		
101	06/01/88	6L'			18700	2670		
101	06/01/88	7A			13400	1650		
101	06/01/88	7A			13400	1580		
101	06/01/88	8A			16300	3650		
101	06/01/88	8A			16300	3620		
102	06/02/88	LL Media			23200	2980		
102	06/02/88	LL Media			23600	2980		
102	06/02/88	LL Media			22800	2760		
103	06/03/88	5L'			17200	2480		
103	06/03/88	5L'			17500	2420		
103	06/03/88	6L'			17800	2510		
103	06/03/88	6L'			17500	2410		
106	06/06/88	5L'	9.35					
106	06/06/88	6L'	9.2					
106	06/06/88	7A		20.33				
106	06/06/88	8A		28.57				
107	06/07/88	5L'			18700	2630		
107	06/07/88	5L'			19500	2750		
107	06/07/88	6L'			19500	2740		
107	06/07/88	6L'			17400	2510		
107	06/07/88	7A			12800	1540		

TABLE D-2
NEW BEDFORD PCB PROJECT
DATA SUMMARY-RADIAN

Day	Date	Reactor Number	D.O. mg/L	O.U. mg/L/hr	T.S. ppm	T.V.S. ppm	TURBIDITY NTU	HANTZSCH FORMALDEHYDE %
107	06/07/88	7A			12800	1550		
107	06/07/88	8A			14900	3120		
107	06/07/88	8A			13900	2120		
108	06/08/88	5L'	11.6	8.11				
108	06/08/88	6L'	11.5	5				
109	06/09/88	5L'			18800	2560		
109	06/09/88	5L'			19100	2600		
109	06/09/88	6L'			18300	2450		
109	06/09/88	6L'			19000	2590		
113	06/13/88	7A		35.7				
113	06/13/88	8A		32.4				
		LOW LEVEL PRESUMPTIVE						
114	06/14/88	7A			14900	2860		
114	06/14/88	7A			15000	2910		
114	06/14/88	8A			16600	3840		
114	06/14/88	8A			16500	3730		
120	06/20/88	5L'		5.33				
120	06/20/88	6L'		6.2				
		LOW LEVEL CONFIRMATION TEST						
129	06/29/88	C5L			21600	2439		
129	06/29/88	C5L			22900	2827		
129	06/29/88	C6L			22600	2858		
129	06/29/88	C6L			21900	2732		
129	06/29/88	8CCL			22600	3447		
129	06/29/88	8CCL			19600	2892		
142	07/12/88	C5L			22100	2883		
142	07/12/88	C5L			22700	2912		
142	07/12/88	C6L			21300	2662		
142	07/12/88	C6L			21100	2688		
142	07/12/88	8CCL			20800	2270		
142	07/12/88	8CCL			21100	2019		

APPENDIX E

GSRI PCB DATA SUMMARY

TABLE E-1. PCB SAMPLE IDENTIFICATION

Source	GSRI No.	Radian No.	
Seed Material			
Intertidal Zone	AK064		
CSO	AK065		
Low Level	AK066		
High Level	AK068		
PCB Reactor	AI024		
Presumptive Phase			
P(INF)H	AK365/366	A8-05-041	01A
P1H	AK371/372		
P2H	AK360/361	A8-05-041	02A
P3H	AK368/369		
P4H	AK363/364	A8-05-041	03A
P(INF)L	AN001	A8-06-071	01A
P5L	AN002	A8-06-071	02A, 02B
P6L	AN003	A8-06-071	03A
Confirmation Phase			
C1H (T=0)	AK362/367		
C2H (T=0)	AK374/375	A8-05-059	01A
C3H (T=0)	AK376/377		
C4H (T=0)	AK378/379		
7CCH (T=0)	AK380/381		
C1H (T=14)	AK382		
C2H (T=14)	AK383	A8-06-008	01A
C3H (T=14)	AK384		
C4H (T=14)	AK385		
7CCH (T=14)	AK386	A8-06-008	02A
C5L (T=0)	AN004	A8-06-083	01A, 02A
C6L (T=0)	AN005		
8CCL (T=0)	AN006		
C5L (T=14)	AK871	A8-07-033	01A, 02B
C6L (T=14)	AK872		
8CCL (T=14)	AK873	A8-07-033	02A

SUMMARY TABLE PROJECT: NEW BEDFORD HARBOR

GSRI

PRE-TEST RESULTS (RADIAN) POST-TEST RESULTS (RADIAN)

SAMPLE LOCATION:		RADIAN-MUD-LL	CSO-AEROBIC-LL	R-LL-AEROBIC	R-HL	U OF W	P2H	P2H	C1H
SMO NUMBER:		AK 064	AK 065	AK 066	AK 068	A1024	AK360	AK361	AK362
UNITS:		ug/kg	ug/kg	ug/kg	ug/kg	ug/l	ug/l	ug/l	ug/l
PESTICIDES/PCB	CRDL ug/kg / CRDL ug/l								
Heptachlor	8 / 0.05	-	-	-	-	22	-	NR	-
Aroclor-1242	80 / 0.50	-	-	1100000	2000000	-	-	NR	-
Aroclor-1248	80 / 0.50	35000	4600	-	-	120	49000 D	NR	86000
Aroclor-1254	160 / 1.00	20000	6800	140000	1300000	-	14000 D	NR	15000
Percent Solids		67	71	34	31	-	-		-
Dilution Factor		55	13	136	109	100	100		100
Dry Wt./Dilution Factor		82	18	400	350	-	-		-
PCB CONGENERS									
Monochlorobiphenyls		-	-	-	-	-	NR	-	NR
Dichlorobiphenyls		-	-	-	1400000	-	NR	1700	NR
Trichlorobiphenyls		-	-	-	3900000	26	NR	20000	NR
Tetrachlorobiphenyls		-	-	-	4600000	91	NR	35600	NR
Pentachlorobiphenyls		-	-	-	2500000	-	NR	22700	NR
Hexachlorobiphenyls		-	-	-	620000	-	NR	6400	NR
Heptachlorobiphenyls		-	-	-	-	-	NR	620	NR
Octachlorobiphenyls		-	-	-	-	-	NR	-	NR
Percent Solids		67	66	38	30	-		-	
Dilution Factor		30	4	30	30	1		100	
Dry Wt./Dilution Factor		1.5	6	79	100	-		-	

SUMMARY TABLE
GERI

SAMPLE LOCATION:		P4H	P4H	P(INF)H	P(INF)H	C1H	C1H	P3H	P3H
SMO NUMBER:		AK363	AK364	AK365	AK366	AK367	AK367DL	AK368	AK369
UNITS:		ug/l	ug/l	ug/l	ug/l	ug/l	ug/l	ug/l	ug/l
PESTICIDES/PCB	CRDL ug/kg	CRDL ug/l							
Heptachlor	8	0.05	-	NR	-	NR	NR	NR	-	NR
Aroclor-1242	80	0.50	-	NR	-	NR	NR	NR	-	NR
Aroclor-1248	80	0.50 D	110000 D	NR	110000 D	NR	NR	NR	82000 D	NR
Aroclor-1254	160	1.00 D	27000 D	NR	18000 D	NR	NR	NR	20000 D	NR
Percent Solids			-		-				-	
Dilution Factor			100		100				100	
Dry Wt./Dilution Factor			-		-				-	
PCB CONGENERS										
Monochlorobiphenyls			NR	-	NR	-	-	-	NR	-
Dichlorobiphenyls			NR	1800	NR	6100 D	3300	3000	NR	2300
Trichlorobiphenyls			NR	22200	NR	33600 D	29900	27000	NR	22700
Tetrachlorobiphenyls			NR	35600	NR	35000 D	35300	32000	NR	34200
Pentachlorobiphenyls			NR	20200	NR	19800 D	20000	17000	NR	20200
Hexachlorobiphenyls			NR	5700	NR	5200 D	5400	4200	NR	5500
Heptachlorobiphenyls			NR	510	NR	500	460	140	NR	490
Octachlorobiphenyls			NR	-	NR	-	-	-	NR	-
Percent Solids				-		-	-	-		-
Dilution Factor				100		100	100	500		100
Dry Wt./Dilution Factor				-		-	-	-		-

SUMMARY TABLE
GERI

SAMPLE LOCATION:			P3HMS P3HMSD	P1H	P1H	P1HMS P1HMSD	C2H	C2H	C3H	C3H
SMO NUMBER:			AK370	AK371	AK372	AK373	AK374	AK375	AK376	AK377
UNITS:			ug/l	ug/l	ug/l	ug/l	ug/l	ug/l	ug/l	ug/l
PESTICIDES/PCB	CRDL ug/kg	CRDL ug/l								
Heptachlor	8	0.05	-	-	NR	NR	-	NR	-	NR
Aroclor-1242	80	0.50	-	-	NR	NR	-	NR	-	NR
Aroclor-1248	80	0.50	86000 D	97000 D	NR	NR	80000 D	NR	100000 D	NR
Aroclor-1254	160	1.00	21000 D	20000 D	NR	NR	13000 D	NR	17000 D	NR
Percent Solids			-	-			-		-	
Dilution Factor			100	100			100		100	
Dry Wt./Dilution Factor			-	-			-		-	
PCB CONGENERS										
Monochlorobiphenyls			NR	NR	-	51	NR	-	NR	-
Dichlorobiphenyls			NR	NR	6200	7400 D	NR	2300	NR	2900
Trichlorobiphenyls			NR	NR	28400 D	33100 D	NR	21800	NR	26000
Tetrachlorobiphenyls			NR	NR	35200 D	41400 D	NR	27300	NR	28000
Pentachlorobiphenyls			NR	NR	19300 D	22700 D	NR	14300	NR	16000
Hexachlorobiphenyls			NR	NR	4600 D	5600	NR	3300	NR	4500
Heptachlorobiphenyls			NR	NR	500	480	NR	200	NR	430
Octachlorobiphenyls			NR	NR	-	-	NR	-	NR	-
Percent Solids					-	-		-		-
Dilution Factor					100	100		100		100
Dry Wt./Dilution Factor					-	-		-		-

SUMMARY TABLE
GERI

	SAMPLE LOCATION:	C4H	C4H
	SMO NUMBER:	AK378	AK379
	UNITS:	ug/l	ug/l
PESTICIDES/PCB	CRDL ug/kg / CRDL ug/l		
Heptachlor	8 / 0.05	-	NR
Aroclor-1242	80 / 0.50	-	NR
Aroclor-1248	80 / 0.50	94000 D	NR
Aroclor-1254	160 / 1.00	15000 D	NR
Percent Solids		-	
Dilution Factor		100	
Dry Wt./Dilution Factor		-	
PCB CONGENERS			
Monochlorobiphenyls		NR	-
Dichlorobiphenyls		NR	2600
Trichlorobiphenyls		NR	24400
Tetrachlorobiphenyls		NR	28400
Pentachlorobiphenyls		NR	15100
Hexachlorobiphenyls		NR	3900
Heptachlorobiphenyls		NR	340
Octachlorobiphenyls		NR	-
Percent Solids			-
Dilution Factor			100
Dry Wt./Dilution Factor			-

SUMMARY TABLE PROJECT: NEW BEDFORD HARBOR

GSRI

SAMPLE LOCATION:				REACTOR C1H	REACTOR C2H	REACTOR C3H	REACTOR C4H	REACTOR 7CCH
SMO NUMBER:	AK 380		AK 381	AK 382	AK 383	AK 384	AK 385	AK386
UNITS:	ug/l		ug/l	ug/l	ug/l	ug/l	ug/l	ug/l
PESTICIDES/PCB	CRDL ug/l							
Alpha-BHC	0.05	R	NR	NR	NR	NR	NR	NR
Beta-BHC	0.05	R	NR	NR	NR	NR	NR	NR
Delta-BHC	0.05	R	NR	NR	NR	NR	NR	NR
Gamma-BHC (Lindane)	0.05	R	NR	NR	NR	NR	NR	NR
Heptachlor	0.05	R	NR	NR	NR	NR	NR	NR
Aldrin	0.05	R	NR	NR	NR	NR	NR	NR
Heptachlor Epoxide	0.05	R	NR	NR	NR	NR	NR	NR
Endosulfan I	0.05	R	NR	NR	NR	NR	NR	NR
Dieldrin	0.10	R	NR	NR	NR	NR	NR	NR
4,4-DDE	0.10	R	NR	NR	NR	NR	NR	NR
Endrin	0.10	R	NR	NR	NR	NR	NR	NR
Endosulfan II	0.10	R	NR	NR	NR	NR	NR	NR
4,4-DDD	0.10	R	NR	NR	NR	NR	NR	NR
Endosulfan Sulfate	0.10	R	NR	NR	NR	NR	NR	NR
4,4'-DDT	0.10	R	NR	NR	NR	NR	NR	NR
Methoxychlor	0.50	R	NR	NR	NR	NR	NR	NR
Endrin Ketone	0.10	R	NR	NR	NR	NR	NR	NR
Alpha-Chlordane	0.50	R	NR	NR	NR	NR	NR	NR
Gamma-Chlordane	0.50	R	NR	NR	NR	NR	NR	NR
Toxaphene	1.00	R	NR	NR	NR	NR	NR	NR
Aroclor-1016	0.50	R	NR	NR	NR	NR	NR	NR
Aroclor-1221	0.50	R	NR	NR	NR	NR	NR	NR
Aroclor-1232	0.50	R	NR	NR	NR	NR	NR	NR
Aroclor-1242	0.50	100000 C	NR	NR	NR	NR	NR	NR
Aroclor-1248	0.50	R	NR	NR	NR	NR	NR	NR
Aroclor-1254	1.00	35000 C	NR	NR	NR	NR	NR	NR
Aroclor-1260	1.00	R	NR	NR	NR	NR	NR	NR
DILUTION FACTOR		2000						
PCB/CONGENERS								
Monochlorobiphenyls		NR	610	-	-	-	-	210
Dichlorobiphenyls		NR	19900 D	540	130	300	-	4100
Trichlorobiphenyls		NR	56900 D	9900	7700	6500	3100	12000
Tetrachlorobiphenyls		NR	54900 D	25600	25300	23000	10700	12200
Pentachlorobiphenyls		NR	26900 D	13100	13800	12800	6700	6500
Hexachlorobiphenyls		NR	6300 D	3200	3600	3300	1900	1800
Heptachlorobiphenyls		NR	460	190	200	180	-	-
DILUTION FACTOR			100	100	100	100	100	100

SUMMARY TABLE PROJECT: NEW BEDFORD HARBOR

GSRI

SAMPLE LOCATION:		AN-001	AN-002	AN-003	AN-004	AN-005	AN-006
SMO NUMBER:							
UNITS:		ug/l	ug/l	ug/l	ug/l	ug/l	ug/l
PESTICIDES/PCB	CRDL ug/l						
Alpha-BHC	0.05	-	-	-	-	R	-
Beta-BHC	0.05	-	-	-	-	R	-
Delta-BHC	0.05	-	-	-	-	R	-
Gamma-BHC (Lindane)	0.05	-	-	-	-	R	-
Heptachlor	0.05	-	-	-	-	R	-
Aldrin	0.05	-	-	-	-	R	-
Heptachlor Epoxide	0.05	-	-	-	-	R	-
Endosulfan I	0.05	-	-	-	-	R	-
Dieldrin	0.10	-	-	-	-	R	-
4,4-DDE	0.10	-	-	-	-	R	-
Endrin	0.10	-	-	-	-	R	-
Endosulfan II	0.10	-	-	-	-	R	-
4,4-DDD	0.10	-	-	-	-	R	-
Endosulfan Sulfate	0.10	-	-	-	-	R	-
4,4'-DDT	0.10	-	-	-	-	R	-
Methoxychlor	0.50	-	-	-	-	R	-
Endrin Ketone	0.10	-	-	-	-	R	-
Alpha-Chlordane	0.50	-	-	-	-	R	-
Gamma-Chlordane	0.50	-	-	-	-	R	-
Toxaphene	1.00	-	-	-	-	R	-
Aroclor-1016	0.50	-	-	-	-	R	-
Aroclor-1221	0.50	-	-	-	-	R	-
Aroclor-1232	0.50	-	-	-	-	R	-
Aroclor-1242	0.50	6900	-	-	-	R	7400
Aroclor-1248	0.50	-	10000	8800	12000	14000	-
Aroclor-1254	1.00	1200	1200	950	1200	1300	910
Aroclor-1260	1.00	-	-	-	-	R	-
DILUTION FACTOR		100	100	100	100	100	100
PCB CONGENERS							
Dichlorobiphenyls		800	170	140	240	180	1700
Trichlorobiphenyls		3100	1600	1600	2000	1700	4300
Tetrachlorobiphenyls		2200	1900	1900	2200	1700	2800
Pentachlorobiphenyls		1700	1600	1600	1900	1500	2100
Hexachlorobiphenyls		390	400	450	540	380	450
DILUTION FACTOR		100	100	100	100	100	100

SUMMARY TABLE PROJECT: NEW BEDFORD HARBOR

GSRI

SAMPLE LOCATION:				
SMO NUMBER:		AK-871	AK-872	AK-873
UNITS:		ug/l	ug/l	ug/l
PESTICIDES/PCB	CRDL ug/l			
Alpha-BHC	0.05	R	-	-
Beta-BHC	0.05	R	-	-
Delta-BHC	0.05	R	-	-
Gamma-BHC (Lindane)	0.05	R	-	-
Heptachlor	0.05	R	-	-
Aldrin	0.05	R	-	-
Heptachlor Epoxide	0.05	R	-	-
Endosulfan I	0.05	R	-	-
Dieldrin	0.10	R	-	-
4,4-DDE	0.10	R	-	-
Endrin	0.10	R	-	-
Endosulfan II	0.10	R	-	-
4,4-DDD	0.10	R	-	-
Endosulfan Sulfate	0.10	R	-	-
4,4'-DDT	0.10	R	-	-
Methoxychlor	0.50	R	-	-
Endrin Ketone	0.10	R	-	-
Alpha-Chlordane	0.50	R	-	-
Gamma-Chlordane	0.50	R	-	-
Toxaphene	1.00	R	-	-
Aroclor-1016	0.50	R	-	-
Aroclor-1221	0.50	R	-	-
Aroclor-1232	0.50	R	-	-
Aroclor-1242	0.50	R	-	7400
Aroclor-1248	0.50	2600	3100	-
Aroclor-1254	1.00	2600	3100	2900
Aroclor-1260	1.00	R	-	-
Dilution Factor		100	100	100
PCB CONGENERS				
Dichlorobiphenyls		120	110	1500
Trichlorobiphenyls		1600	1600	4200
Tetrachlorobiphenyls		2500	2500	2700
Pentachlorobiphenyls		1800	1900	1900
Hexachlorobiphenyls		400	580	480
Dilution Factor		100	100	100

NOTE: AK 873 - difficult to distinguish between 1242 & 1248 when 1254 is present.

APPENDIX F

RADIAN PCB DATA SUMMARY

TABLE F-1. PCB SAMPLE IDENTIFICATION

Source	GSRI No.	Radian No.	
Seed Material			
Intertidal Zone	AK064		
CSO	AK065		
Low Level	AK066		
High Level	AK068		
PCB Reactor	AI024		
Presumptive Phase			
P(INF)H	AK365/366	A8-05-041	01A
P1H	AK371/372		
P2H	AK360/361	A8-05-041	02A
P3H	AK368/369		
P4H	AK363/364	A8-05-041	03A
P(INF)L	AN001	A8-06-071	01A
P5L	AN002	A8-06-071	02A, 02B
P6L	AN003	A8-06-071	03A
Confirmation Phase			
C1H (T=0)	AK362/367		
C2H (T=0)	AK374/375	A8-05-059	01A
C3H (T=0)	AK376/377		
C4H (T=0)	AK378/379		
7CCH (T=0)	AK380/381		
C1H (T=14)	AK382		
C2H (T=14)	AK383	A8-06-008	01A
C3H (T=14)	AK384		
C4H (T=14)	AK385		
7CCH (T=14)	AK386	A8-06-008	02A
C5L (T=0)	AN004	A8-06-083	01A, 02A
C6L (T=0)	AN005		
8CCL (T=0)	AN006		
C5L (T=14)	AK871	A8-07-033	01A, 02B
C6L (T=14)	AK872		
8CCL (T=14)	AK873	A8-07-033	02A

PCB ANALYSIS (EPA METHOD 680)
RADIAN CORPORATION

NEW BEDFORD HARBOR SEDIMENTS

Lab No:	A8-05-041	A8-05-041	A8-05-041
Sample No:	01A	02A	03A
Description:	P(INF)H	P2H	P3H
Isomer Group			
Monochlorobiphenyls	3.2	<2.0	<2.0
Dichlorobiphenyls	790	290	220
Trichlorobiphenyls	3700	5200	4300
Tetrachlorobiphenyls	3700	9000	720
Pentachlorobiphenyls	1100	2900	2200
Hexachlorobiphenyls	620	1700	1200
Heptachlorobiphenyls	80	230	160
Octachlorobiphenyls	11	32	22
Nonachlorobiphenyls	<2.0	4.2	3.3
Decachlorobiphenyls	<2.0	<2.0	<2.0

Lab No:	A8-06-071	A8-06-071	A8-06-071
Sample No:	01A	02A	03A
Description:	P(INF)L	P5L	P5L-DUP
Isomer Group			
Monochlorobiphenyls	<0.8	<0.7	0.7*
Dichlorobiphenyls	430	120	150
Trichlorobiphenyls	1800	1200	1400
Tetrachlorobiphenyls	1600	1800	2100
Pentachlorobiphenyls	600	670	780
Hexachlorobiphenyls	270	320	360
Heptachlorobiphenyls	30	40	46
Octachlorobiphenyls	<4.9	<4.3	7.5*
Nonachlorobiphenyls	<7.9	<6.9	<6.4
Decachlorobiphenyls	<9.1	<8.0	<7.4

*Less than 5 times the detection limit.

All units are ng/ml unless noted.

PCB ANALYSIS (EPA METHOD 680)
RADIAN CORPORATION

NEW BEDFORD HARBOR SEDIMENTS

Lab No:	A8-06-071	A8-06-071	A8-05-059
Sample No:	03A	04A	01A
Description:	P6L	Reagent Blank	C2H(T=0)
Isomer Group			
Monochlorobiphenyls	<0.8	<0.7	<2.0
Dichlorobiphenyls	69	<0.6	510
Trichlorobiphenyls	1000	<1.3	6900
Tetrachlorobiphenyls	1700	<2.0	8300
Pentachlorobiphenyls	660	<1.4	2400
Hexachlorobiphenyls	310	<2.5	1300
Heptachlorobiphenyls	38	<3.5	160
Octachlorobiphenyls	<4.8	<4.2	21
Nonachlorobiphenyls	<7.7	<6.8	2.8
Decachlorobiphenyls	<8.9	<7.8	<2.0

Lab No:	A8-05-059	A8-06-008**	A8-06-008**
Sample No:	02A	01A	02A
Description:	Reagent Blank	C2H(T=14)	7CCH(T=14)
Isomer Group			
Monochlorobiphenyls	<2.0	4.2*	650
Dichlorobiphenyls	<2.0	400	11000
Trichlorobiphenyls	<2.0	7200	45000
Tetrachlorobiphenyls	<2.0	30000	53000
Pentachlorobiphenyls	<2.0	7300	17000
Hexachlorobiphenyls	<2.0	3300	8200
Heptachlorobiphenyls	<2.0	410	1100
Octachlorobiphenyls	<2.0	54	160
Nonachlorobiphenyls	<2.0	19	60
Decachlorobiphenyls	<2.0	<2.0	<2.0

*Less than 5 times the detection limit.
All units are ng/ml unless noted.
**ug/l

PCB ANALYSIS (EPA METHOD 680)
RADIAN CORPORATION

NEW BEDFORD HARBOR SEDIMENTS

Lab No:	A8-06-008	A8-07-033	A8-07-033
Sample No:	03A	01A	02B
Description:	Reagent Blank	C5L(T=14)	C5L-DUP(T=14)
Isomer Group			
Monochlorobiphenyls	<2.0	<0.6	<0.7
Dichlorobiphenyls	<2.0	41	57
Trichlorobiphenyls	<2.0	610	840
Tetrachlorobiphenyls	<2.0	1300	1900
Pentachlorobiphenyls	<2.0	530	750
Hexachlorobiphenyls	<2.0	250	360
Heptachlorobiphenyls	<2.0	28	42
Octachlorobiphenyls	<2.0	4.6*	6.3*
Nonachlorobiphenyls	<2.0	<5.6	<6.3
Decachlorobiphenyls	<2.0	<6.9	<7.8

Lab No:	A8-07-033	A8-07-033	A8-06-083
Sample No:	02A	03A	01A
Description:	8CCL(T=14)	Reagent Blank	C5L(T=0)
Isomer Group			
Monochlorobiphenyls	3.6	<0.8	<0.7
Dichlorobiphenyls	430	<0.7	85
Trichlorobiphenyls	1900	<1.6	1100
Tetrachlorobiphenyls	1900	<2.2	1900
Pentachlorobiphenyls	740	<1.5	730
Hexachlorobiphenyls	360	<2.9	350
Heptachlorobiphenyls	42	<3.8	42
Octachlorobiphenyls	7.1*	<4.6	6.5*
Nonachlorobiphenyls	<5.9	<6.9	<5.9
Decachlorobiphenyls	<7.3	<8.6	<7.2

*Less than 5 times the detection limit.

All units are ng/ml unless noted.

PCB ANALYSIS (EPA METHOD 680)
RADIAN CORPORATION

NEW BEDFORD HARBOR SEDIMENTS

Lab No:	A8-06-083	A8-06-083
Sample No:	02A	03A
Description:	C5L-DUP(T=0)	Reagent Blank
Isomer Group		
Monochlorobiphenyls	<0.8	<0.5
Dichlorobiphenyls	110	<0.5
Trichlorobiphenyls	1400	<1.1
Tetrachlorobiphenyls	2200	<1.5
Pentachlorobiphenyls	860	<1.0
Hexachlorobiphenyls	410	<1.9
Heptachlorobiphenyls	49	<2.6
Octachlorobiphenyls	5.1*	<3.1
Nonachlorobiphenyls	<6.9	<4.6
Decachlorobiphenyls	<8.4	<5.7

All units are ng/ml unless noted.

APPENDIX G

RADIAN PCB ANALYSIS QUALITY CONTROL DATA

RADIAN

Client: EBASCO
RADIAN MILWAUKEE
5103 W. BELOIT RD.
MILWAUKEE, WI. 53214

01A P (INF) H
02A P 2 H
03A P 4 H

EPA METHOD 680
RESULTS IN ng/mL
Lab No: A8-05-041

CAS #	COMPOUND	01A	02A	03A
	Monochlorobiphenyls	3.2	<2.0	<2.0
	Dichlorobiphenyls	790	290	220
	Trichlorobiphenyls	3700	5200	4300
	Tetrachlorobiphenyls	3700	9000	720
	Pentachlorobiphenyls	1100	2900	2200
	Hexachlorobiphenyls	620	1700	1200
	Heptachlorobiphenyls	80	230	160
	Octachlorobiphenyls	11	32	22
	Nonachlorobiphenyls	<2.0	4.2	3.3
	Decachlorobiphenyls	<2.0	<2.0	<2.0

SURROGATE RECOVERIES (results in % recovery)

Monochlorobiphenyls	56	67	54
Tetrachlorobiphenyls	88	89	86
Octachlorobiphenyls	95	96	89
Decachlorobiphenyls	94	76	90

RADIAN

Client: EBASCO
RADIAN MILWAUKEE
5103 W. BELOIT RD.
MILWAUKEE, WI. 53214

01A REACTOR C2H (INITIAL COND)
02A REAGENT BLANK

EPA METHOD 680
RESULTS IN ng/mL
Lab No: A8-05-059

CAS #	COMPOUND	01A	02A
	Monochlorobiphenyls	<2.0	<2.0
	Dichlorobiphenyls	510	<2.0
	Trichlorobiphenyls	6900	<2.0
	Tetrachlorobiphenyls	8300	<2.0
	Pentachlorobiphenyls	2400	<2.0
	Hexachlorobiphenyls	1300	<2.0
	Heptachlorobiphenyls	160	<2.0
	Octachlorobiphenyls	21	<2.0
	Nonachlorobiphenyls	2.8	<2.0
	Decachlorobiphenyls	<2.0	<2.0

SURROGATE RECOVERIES	(results in % recovery)	
Monochlorobiphenyls	49	50
Tetrachlorobiphenyls	84	78
Octachlorobiphenyls	91	79
Decachlorobiphenyls	67	54

RADIAN
CORPORATION

Page 1

Client: EBASCO
RADIAN MILWAUKEE
5103 W. BELOIT RD.
MILWAUKEE, WI. 53214

01A REACTOR C 2H
02A REACTOR 7C CH
03A REAGENT BLANK

EPA METHOD 680

Lab No: 88-06-008

RESULTS IN ug/L

CAS #	COMPOUND	01A	02A	03A
	Monochlorobiphenyls	4.2*	650	<2.0
	Dichlorobiphenyls	490	11000	<2.0
	Trichlorobiphenyls	7200	45000	<2.0
	Tetrachlorobiphenyls	30000	53000	<2.0
	Pentachlorobiphenyls	7300	17000	<2.0
	Hexachlorobiphenyls	3300	8200	<2.0
	Heptachlorobiphenyls	410	1100	<2.0
	Octachlorobiphenyls	54	160	<2.0
	Nonachlorobiphenyls	19	60	<2.0
	Decachlorobiphenyls	<2.0	<2.0	<2.0

SURROGATE RECOVERIES (results in % recovery)

Monochlorobiphenyls	86	128	61
Tetrachlorobiphenyls	115	146	89
Octachlorobiphenyls	109	141	77
Decachlorobiphenyls	99	113	65

NOTES AND DEFINITIONS FOR THIS REPORT.
QC = OUTSIDE CONTROL LIMITS.
* = LESS THAN 5 TIMES THE DETECTION LIMIT.
B = DETECTED IN REAGENT BLANK; BACKGROUND SUBTRACTION NOT PERFORMED.
ND = NOT DETECTED AT DETECTION LIMIT.
NA = NOT ANALYZED.
N\A = NOT AVAILABLE.
NS = NOT SPIKED.

Rec. 6/15/85

RADIAN

Client: EBASCO
RADIAN MILWAUKEE
5103 W. BELOIT RD.
MILWAUKEE, WI. 53214

EPA METHOD 680

01A Influent low level comp.
02A Effluent 5L composite
02B Effluent 5L comp. dup.
03A Effluent 6L composite

Lab No: A8-06-071

RESULTS IN ng/ml

CAS #	COMPOUND	01A	02A	02B	03A
	Monochlorobiphenyls	<0.8	<0.7	0.7*	<0.8
	Dichlorobiphenyls	430	120	150	69
	Trichlorobiphenyls	1800	1200	1400	1000
	Tetrachlorobiphenyls	1600	1800	2100	1700
	Pentachlorobiphenyls	600	670	780	660
	Hexachlorobiphenyls	270	320	360	310
	Heptachlorobiphenyls	30	40	46	38
	Octachlorobiphenyls	<4.9	<4.3	7.5*	<4.8
	Nonachlorobiphenyls	<7.9	<6.9	<6.4	<7.7
	Decachlorobiphenyls	<9.1	<8.0	<7.4	<8.9

SURROGATE RECOVERIES (results in % recovery)

Monochlorobiphenyls	55	22QC	41QC	50
Tetrachlorobiphenyls	66	78	80	75
Octachlorobiphenyls	70	84	85	79
Decachlorobiphenyls	69	90	73	85

NOTES AND DEFINITIONS FOR THIS REPORT.
QC = OUTSIDE CONTROL LIMITS.
* = LESS THAN 5 TIMES THE DETECTION LIMIT.
B = DETECTED IN REAGENT BLANK; BACKGROUND SUBTRACTION NOT PERFORMED.
ND = NOT DETECTED AT DETECTION LIMIT.
NA = NOT ANALYZED.
N\A = NOT AVAILABLE.
NS = NOT SPIKED.

RADIAN

04A Reagent Blank

Client: EBASCO
RADIAN MILWAUKEE
5103 W. BELOIT RD.
MILWAUKEE, WI. 53214

EPA METHOD 680

Lab No: A8-06-071

RESULTS IN ng/ml

CAS #	COMPOUND	04A
	Monochlorobiphenyls	<0.7
	Dichlorobiphenyls	<0.6
	Trichlorobiphenyls	<1.3
	Tetrachlorobiphenyls	<2.0
	Pentachlorobiphenyls	<1.4
	Hexachlorobiphenyls	<2.5
	Heptachlorobiphenyls	<3.5
	Octachlorobiphenyls	<4.2
	Nonachlorobiphenyls	<6.8
	Decachlorobiphenyls	<7.8

SURROGATE RECOVERIES	(results in % recovery)
Monochlorobiphenyls	57
Tetrachlorobiphenyls	76
Octachlorobiphenyls	86
Decachlorobiphenyls	78

NOTES AND DEFINITIONS FOR THIS REPORT.
QC = OUTSIDE CONTROL LIMITS.
* = LESS THAN 5 TIMES THE DETECTION LIMIT.
B = DETECTED IN REAGENT BLANK; BACKGROUND SUBTRACTION NOT PERFORMED.
ND = NOT DETECTED AT DETECTION LIMIT.
NA = NOT ANALYZED.
N\A = NOT AVAILABLE.
NS = NOT SPIKED.

RADIAN

01A Reactor C5L-initial cond.
02A Reactor C5L-init cond. dup
03A Reagent Blank

Client: EBASCO
RADIAN MILWAUKEE
5103 W. BELOIT RD.
MILWAUKEE, WI. 53214

EPA METHOD 680

Lab No: A8-06-083

RESULTS IN ng/ml

CAS #	COMPOUND	01A	02A	03A
	Monochlorobiphenyls	<0.7	<0.8	<0.5
	Dichlorobiphenyls	85	110	<0.5
	Trichlorobiphenyls	1100	1400	<1.1
	Tetrachlorobiphenyls	1900	2200	<1.5
	Pentachlorobiphenyls	730	860	<1.0
	Hexachlorobiphenyls	350	410	<1.9
	Heptachlorobiphenyls	42	49	<2.6
	Octachlorobiphenyls	6.5*	5.1*	<3.1
	Nonachlorobiphenyls	<5.9	<6.9	<4.6
	Decachlorobiphenyls	<7.2	<8.4	<5.7

SURROGATE RECOVERIES (results in % recovery)

Monochlorobiphenyls	44QC	42QC	69
Tetrachlorobiphenyls	90	97	103
Octachlorobiphenyls	90	95	101
Decachlorobiphenyls	98	102	105

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RADIAN

Client: EBASCO
RADIAN MILWAUKEE
5103 W. BELOIT RD.
MILWAUKEE, WI. 53214
EPA METHOD 680

01A Effluent C5L
02A Effluent 8CCL
02B Effluent C5L dup
03A Reagent Blank

Lab No: A8-07-033

RESULTS IN ng/ml

CAS #	COMPOUND	01A	02A	02B	03A
	Monochlorobiphenyls	<0.6	3.6	<0.7	<0.8
	Dichlorobiphenyls	41	430	57	<0.7
	Trichlorobiphenyls	610	1900	840	<1.6
	Tetrachlorobiphenyls	1300	1900	1900	<2.2
	Pentachlorobiphenyls	530	740	750	<1.5
	Hexachlorobiphenyls	250	360	360	<2.9
	Heptachlorobiphenyls	28	42	42	<3.8
	Octachlorobiphenyls	4.6*	7.1*	6.3*	<4.6
	Nonachlorobiphenyls	<5.6	<5.9	<6.3	<6.9
	Decachlorobiphenyls	<6.9	<7.3	<7.8	<8.6

	SURROGATE RECOVERIES (results in % recovery)			
Monochlorobiphenyls	51	81	69	72
Tetrachlorobiphenyls	65	100	90	101
Octachlorobiphenyls	68	99	96	103
Decachlorobiphenyls	71	97	99	100

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